

Field Demonstration of the Performance of Wastewater Treatment Solution (WTS®) to Reduce Phosphorus and other Substances from Dairy Lagoon Effluent

Final Report August 2008

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Funded by the
Texas State Soil and Water Conservation Board
under CWA Section 319, EPA
TSSWCB Project # 03-10

Partners:
Texas AgriLife Extension Service
Texas Water Resources Institute
Ozona Environmental® LLC, Ozona, Texas

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Summary

In 1998 two upper North Bosque River segments were designated as impaired due to point source and nonpoint source (NPS) pollution of phosphorus (P) in these segments of the watershed. As a result, two Total Maximum Daily Loads (TMDLs) were applied, which called for the reduction of annual loading and annual average soluble reactive P (SRP) concentrations by about 50%. Under the Clean Water Act (Section 319(h)), a new technologies demonstration project was funded by the U. S. Environmental Protection Agency (USEPA) Region 6 and administered by the Texas State Soil and Water Conservation Board (TSSWCB) for reducing water pollution associated with dairy animal production systems. As part of this demonstration, the efficacy of a prospective new technology (i.e. wastewater treatment solution, WTS[®]) was evaluated, which may assist dairy farmers in reducing P from lagoon effluent. In many cases, this effluent is applied to waste application fields (WAF) as irrigation water. Therefore, reducing P in the effluent can have a direct impact on NPS pollution in the watershed.

Before treating a dairy's anaerobic lagoon with WTS[®] and an oxygenating additive, O2T, three separate background (pre-treatment) samplings were conducted to gather baseline information on nutrients (e.g., total phosphorus [TP], soluble reactive phosphorus [SRP], and total Kjeldahl nitrogen [TKN]) and solids (e.g., total solids [TS], total suspended solids [TSS], total dissolved solids [TDS]) data prior to inoculation. Following the third pre-treatment sampling in September 2007, the anaerobic lagoon was treated with WTS[®] at an averaged application rate of 1 gallon/head as a start-up. Thereafter, WTS[®] was applied at a rate of 0.5 gal/100 head-day (based on 600 heads), while O2T was applied at a rate of 0.1 gal/100 head-day (based on 600 heads). To mimic the repeatability of lagoon treatment, two large tanks were filled with untreated flushed manure to assess the treatment effect on flushed manure from free-stall. Tank 1 (T1) was treated manually on a monthly basis, with WTS[®] at a rate of 16 oz (0.5 L) and with O2T at a rate of 7 oz (0.25 L) and Tank 2 (T2) was used as the control (no treatment was applied).

Following treatment, lagoon samples were collected monthly or bi-monthly from two different profiles: lagoon supernatant (LS), sampled from the top of the liquid level to 2 ft (0.61 m) depth and lagoon profile (LP), sampled from the entire depth of the lagoon using a sludge judge (a

sampling tube with a check valve at the bottom to take lagoon sample at different depths). For each LP and LS, 27 samples (3 samples per location \times 9 locations) were collected during each sampling event. A set of 9 LP and 9 LS samples were mixed separately to get two composites of each for nutrients including P, solids, pH, conductivity and metals. Similarly, samples were collected from tank supernatant (1 ft or 0.30 m below liquid surface) and profile (from the entire depth of the tank) in each sampling event. During each sampling event, a total 36 (9 samples per tank \times 2 tanks \times 2 profiles) samples were collected from the two tanks. Each set of 9 tank supernatant and 9 tank profile sample bottles were mixed separately to get two tank supernatant (T1S and T2S) and two tank profile (T1P and T2P) composite samples of each for analysis.

WTS[®] treatment was somewhat effective in reducing sludge depth by 10% compared to its pre-treatment level. This reduction of sludge depth was due to microbial treatment, which will likely improve lagoon effluent characteristics, increase lagoon capacity and reduce maintenance cost for this lagoon. This treatment system increases pH in the LS significantly as compared to LP. Similar to lagoon pH, the treated tank T1 had a slightly higher pH as compared to untreated tank T2 in both tank profiles, although differences were not statistically significant. There was no significant reduction in TS either in lagoon or tank environments due to WTS[®] treatment. Overall TSS was reduced by 7% and 9% for LP and LS, respectively, when concentrations of these parameters averaged across post-treatment events were compared with the averages across pre-treatment events. There were no differences in TSS concentrations of treated and untreated tank samples at either LS or LP. Following microbial treatment of the lagoon, TDS concentration both in LS and LP increased, although no significant differences were observed between the two profiles. Overall, the TDS concentration in LS was 13% higher than that of LP.

There was not a significant reduction in TP in either lagoon sampling profile. TP concentration in the treated tank profile was reduced by 17%, yet increased by 2% in the untreated tank profile samples. However, TP reduction values for treated and untreated tank supernatant samples were 60 and 55%, respectively. This suggested that the differences in TP reduction between treated and untreated samples were due to treatment effects. SRP concentration in both LP and LS samples increased gradually, although differences were not significant between LP and LS. A similar SRP increasing trend was also observed for tank samples, but differed in that the treated

tank had a higher SRP concentration than that of untreated tank samples, due to greater TDS in tank supernatant. TKN in LP and LS reduced by 29 and 19%, respectively, but a greater TKN reduction was observed in tank profile (60 and 47% in treated and untreated tank profile samples, respectively) and tank supernatant samples (88 to 86% in treated and untreated tank supernatant samples, respectively) as compared to lagoon samples. Following the microbial treatment, the conductivity and potassium (K) concentration increased in both profiles of the lagoon and treated tank (T2). Three chemical quality parameters indicate the effectiveness of a wastewater treatment system such as biological oxygen demand (BOD), suspended solids, and TP (van Loon and Duffy 2000). Suspended solids and TP were both monitored in this study and had insignificant variation between pre-treatment and post-treatment. The purpose of this study was to evaluate the effectiveness of WTS[®] in reducing P and other substances from lagoon effluent to be applied to WAFs. Therefore, this treatment system was not very effective in reducing phosphorus and other nutrients from the lagoon effluent, especially soluble parameters. Conclusions indicate that more studies are needed to assess the effectiveness of this treatment over a longer time period.

Introduction

The bulk of the manure from animal feeding operations (AFOs) in the United States is applied to crop and pastureland because it is an excellent resource for plant nutrients and soil conditioning. Excessive land application rates and improper uses of manure, however, can lead to environmental concerns and problems. Manure phosphorus (P) not used by plants represents one concern that can significantly impact surface water quality. Water quality degradation due to nonpoint source P contribution from effluent and manure applied to waste application fields (WAFs) is a major concern in the Bosque River watershed. In 1998, two upper North Bosque River segments (that is, 1255 and 1226) were designated as impaired segments on the Texas Clean Water Act, Section 303(d) list (TNRCC 2001). This designation was the result of excessive nutrient loading and aquatic plant growth in those segments. The changes in the status of the Bosque River segments prompted the Texas Commission on Environmental Quality (TCEQ) to develop Total Maximum Daily Loads (TMDLs) that address P loading in these designated segments. In December 2002, TCEQ approved the implementation plan for these TMDLs; these plans were also approved by the Texas State Soil and Water Conservation Board (TSSWCB) in January 2003. The TMDLs call for a reduction of the annual loading and annual average soluble reactive phosphorus (SRP) concentrations by about 50%.

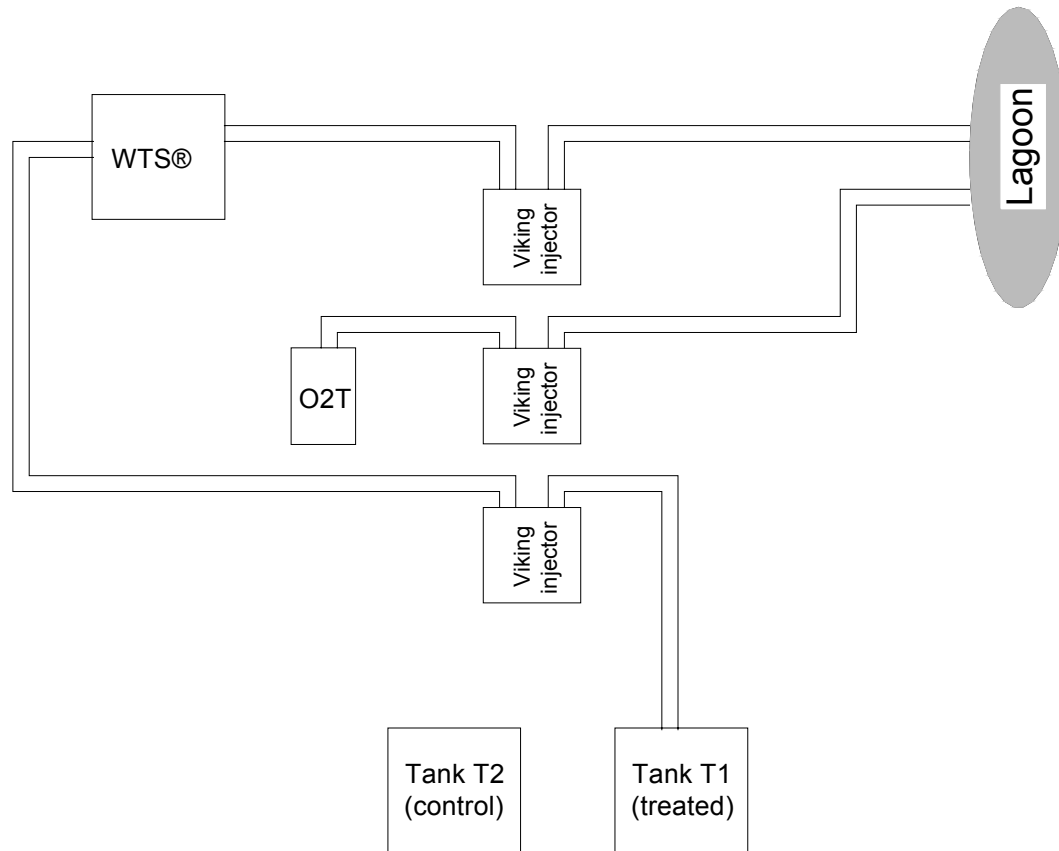
The TCEQ has cited pollution from nonpoint source agricultural operations (by way of runoff) as the main source of contamination to these segments. As a result, reducing P from dairy effluent applied to WAFs is a vital step in protecting the quality of these waterbodies. Runoff from WAFs is not currently regulated as a point source, but its impact on waterbodies can be minimized by using on-farm management practices to reduce potential pollutants in the dairy lagoon effluent prior to WAF application. Currently, a number of dairy operations in the watershed have been using best management practices (BMPs) for removing P and SRP from the wastewater. However, to meet the goals of the established TMDLs, new, more effective and efficient BMPs are needed. One prospective BMP is the use of wastewater treatment solution (WTS[®]) microbial treatment to remove P and other constituents from the effluent stored and treated in dairy lagoons.

This report outlines the performance of a patented liquid-borne WTS[®] introduced by Ozona Environmental[®] LLC, Ozona, Texas. The demonstration evaluated under this project was set up to treat a primary anaerobic dairy lagoon, which has 600-head lactating cows in a free-stall dairy in the Bosque River watershed. Free-stall alleys were flushed twice a day and scraped in the remaining time. As needed, effluent from the lagoon was used to irrigate nearby cropland at the dairy operation using a big gun irrigation system.

Wastewater treatment solution (WTS[®]) treatment system

The WTS[®] treatment system consisted of two parts: a microbial stimulant (WTS[®]) and an oxygenating (O2T) additive (Fig. 1), applied to the lagoon simultaneously. According to the technology provider, microbial treatment systems introduce and stimulate indigenous populations of microorganism, resulting in reduced organic matter and nutrients in the wastewater. The O2T additive provides oxygen to the wastewater to accelerate microbial activity.

In the lagoon, 1 gallon/head of WTS[®] was applied directly to the lagoon in the initial inoculation; thereafter, WTS[®] was applied at a rate of 0.5 gal/100 head-day (based on 600 heads), while O2T was applied at a rate of 0.1 gal/100 head-day (based on 600 heads). A schematic of the WTS[®] treatment system is presented in Fig. 1. As shown in the schematic, two Viking injectors (Viking injector, Kyjac Inc., Pa.) were used for controlling flow rates of WTS[®] and O2T in the lagoon. One additional Viking injector was used to control WTS[®] flow in the treated tank at a predefined rate and interval. This whole system was powered by alkaline lantern batteries.



**Figure 1. Schematic of WTS® treatment system for an anaerobic lagoon and tank
(drawing not to scale)**

Additionally, to mimic the repeatability of lagoon treatment, two large tanks (volume of liquid in Tank 1 [T1] and Tank 2 [T2] was 267 gal (1011 L) and 279 gal (1057 L), respectively) were filled with untreated flushed manure to assess the WTS® treatment effect on flushed manure from free-stall (Fig. 2). Tank T1 was treated manually once a month with 16 oz (0.5 L) of WTS® and 7 oz (0.25L) of O2T. Tank T2 was used as the control (no treatment was applied). To minimize evaporation losses from both tanks, shade cloth covered both tanks and no water was added to compensate for evaporation losses.



Figure 2. Treated tank T1 and control tank T2 used in this study.

Methods

Layout of sampling scheme

Prior to sampling, the lagoon was divided into three, roughly equal, sections by transect lines running along the width and length of the lagoon. The location of each transect was marked with a float and supported by a weight anchored to the float (Fig. 3a). Each intersection was marked and noted as sampling location 1 through 9 (Fig. 3b). In addition, the 10th sampling location was chosen near the flush water inlet (Fig. 3b). A summary of sampling events is listed in Table 1.



Figure 3a. White floats indicating sampling location.

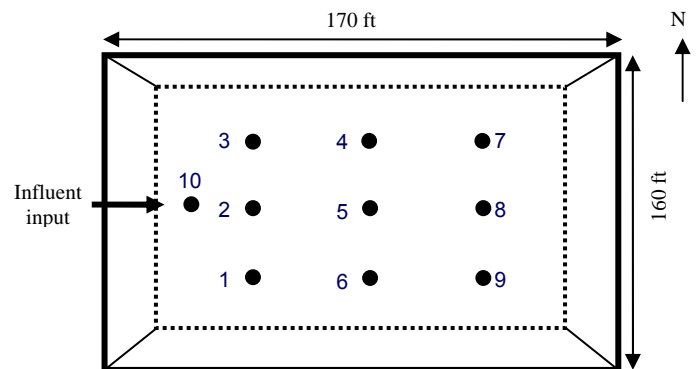


Figure 3b. Schematic of lagoon sampling layout (not to scale).

● Indicates lagoon sampling and sludge depth measurement locations (not to scale).

At each lagoon sampling location, three lagoon supernatant (from top of the liquid level, LS hereafter) and three lagoon profile (from the entire depth of the lagoon, LP hereafter) samples were taken (Fig. 4) for subsequent analysis.

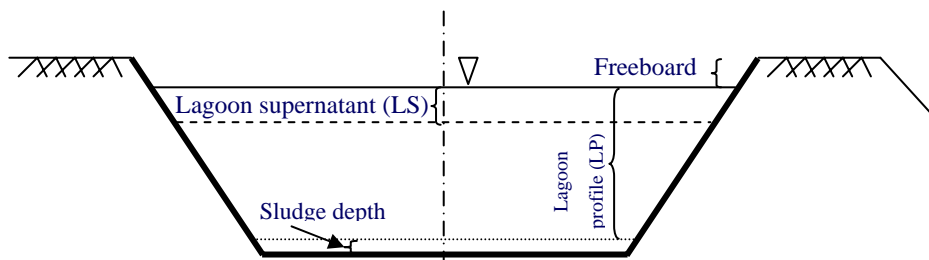


Figure 4. Schematic of lagoon and sampling profile (not to scale).

Component/Date	July, 07	Aug, 07	Sep, 07 ^[a]	Oct, 07	Nov, 07	Jan, 08	Mar, 08
	Pre-treatment sampling			Post-treatment sampling			
Lagoon profile (LP)	√	√	√	√	√	√	√
Lagoon supernatant (LS)	√	√	√	√	√	√	√
Tank supernatant			√*	√	√	√	√
Tank profile			√*	√	√	√	√

Table 1. Sampling events

* Tanks were filled with flushed water and pre-treatment samples were collected from both control and treated tanks.

^[a.] Following pre-treatment samples, treatment begins for both lagoon and tanks.

Similarly, tank samples were also collected from tank supernatant from top of the liquid level to 1 ft (30 cm) depth, and tank profile from the entire depth of the tank in each sampling event as shown in Fig. 5.

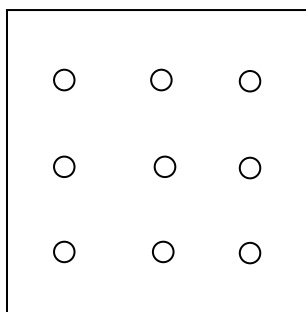


Figure 5. Approximate tank sampling location

Sludge depth (SD) measurement

Typically, reduction of total suspended solids (TSS) in lagoon supernatant is accompanied by reduction of P and a potential change in sludge depth. Therefore, accurate tracking of sludge depth is important to evaluate the performance of WTS[®] treatment effectively. During each sampling event, total depth (TD) and the depth above dense sludge (DADS) for the lagoon and tanks were measured using a measuring tape tied to a metal conduit fitted with an end cap (Fig. 6a). All depth measurements in the lagoon were taken at the same location as liquid samples were collected. Sludge depth (SD) of lagoon and tanks was estimated by subtracting the DADS from the TD of the lagoon and tanks, respectively.

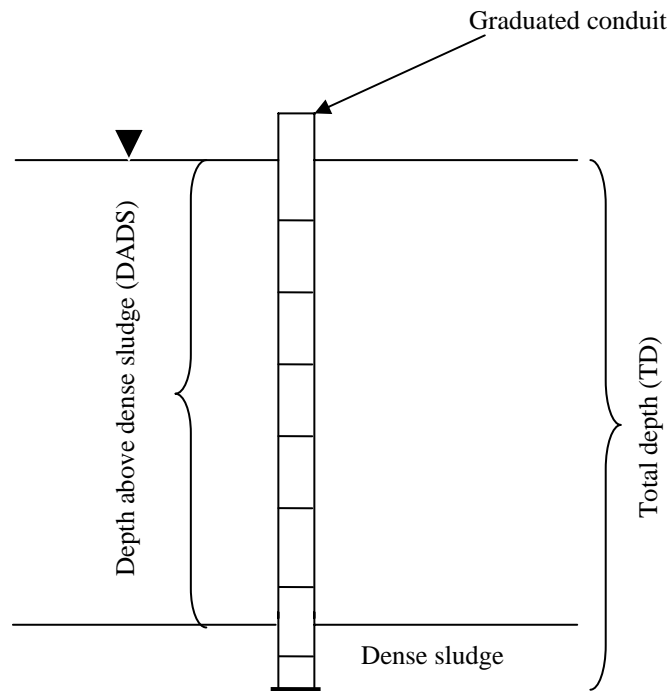


Figure 6a. Schematic of lagoon depth measurement.



Figure 6b. Actual depth measurement using a graduated scale attached to a solid conduit with a flat bottom.

Lagoon and tank effluent sample collection

In order to ensure consistent sampling and monitoring, lagoon sampling locations and the sampling profile were predetermined (Figs. 3b & 4). Before treating the lagoon with WTS[®] and O2T, three background (pre-treatment) samples were taken as described in Table 1 to gather baseline information on nutrients (total phosphorus [TP], SRP, and total Kjeldahl nitrogen [TKN]) and solids data (total solids [TS], TSS, and total dissolved solids [TDS]). For each of the first two pre-treatment sampling events (July and August 2007), 9 composite samples were collected in each sampling event and analyzed (one composite sample from each location as shown in Fig. 3b). Samples were collected using the “Ultra Sludge Judge” (Nasco, Fort Atkinson, WI), which consisted of three 5 ft (1.52 m) sections of 1.25 inch (0.03 m) diameter acrylic tube and a ball check valve at the bottom end (Fig. 7). For LS sampling, the sludge sampler was lowered slowly to the desired depth (2 ft, or 0.61 m), while for LP sampling, the sampler was lowered slowly until it rested above the dense sludge at the bottom of lagoon. After lowering the sludge sampler at desired depth, it was gently pulled out of lagoon as straight as possible.

Based on the first two pre-treatment sample analysis results, all LS and LP samples were divided into three groups (group1: locations 1-3, group 2: locations 4-6, and group 3: locations 7-9). For

subsequent pre- and post-treatment sampling, three LS and three LP samples were taken from each location within a group. A total of 27 LS (3 samples per location \times 9 locations) and 27 LP (3 samples per location \times 9 locations) samples were collected from the lagoon during each sampling event. Sample preparation and analysis for LS and LP will be discussed in the following section.



Figure 7. Lagoon sampling using a sludge judge.

Following the lagoon sampling procedures, 9 tank supernatant and 9 tank profile samples were collected from each tank using sludge sampler (Fig. 8). Thus, 36 (9 samples per tank \times 2 tanks \times 2 profiles) samples were collected from two tanks during each sampling event. Sample preparation and analysis for tank supernatant and tank profile will be discussed in the following section.



Figure 8. Tank sampling using a sludge judge.

Within an hour of conducting sampling, bottles kept on ice were transported to Texas Institute for Applied Environmental Research (TIAER) laboratory at Tarleton State University in Stephenville, Texas, for physicochemical parameters analysis (i.e., nutrients, solids, metals, pH and conductivity).

Sample preparation and analysis

After each sampling event, 9 LS samples were mixed together to obtain one LS composite sample. Similarly, 9 LP samples were mixed together to obtain one LP composite sample. In this way, three LS and three LP composite samples (LS1 & LP1 composited samples from group 1, LS2 & LP2 composited samples from group 2, and LS3 & LP3 composited samples from group 3) were prepared for analysis. Similarly, each set of 9 tank supernatant and 9 tank profile sample bottles were mixed separately to get two tank supernatant (T1S and T2S) and two tank profile (T1P and T2P) composite samples of each for analysis.

Using EPA laboratory procedures (Budde, 1995) and Standard Methods (APHA, 2005) (Table 2), all composited samples were analyzed for: TS, total volatile solids (TVS), total fixed solids (TFS), TSS, SRP, TP, nitrate/nitrite-nitrogen (NNN), TKN,, potassium (K), aluminum (Al),

calcium (Ca), magnesium (Mg), sodium (Na), manganese (Mn), iron (Fe), and Copper (Cu). Concentrations of TDS were found by subtracting the concentrations of TSS from TS. Also pH and conductivity were measured for each composite sample.

Table 2. Laboratory analytical methods

Parameter	Method	Equipment Used
Nitrite + +Nitrate Nitrogen	EPA 353.2 and SSSA 38-1148	Perstorp® or Lachat® QuickChem Autoanalyzer
Total Kjeldahl Nitrogen (TKN)	EPA 353.2, modified	Perstorp® or Lachat® QuickChem Autoanalyzer
Potassium	EPA 200.7	Spectro ® ICP
Calcium	EPA 200.7	Spectro ® ICP
Magnesium	EPA 200.7	Spectro ® ICP
Sodium	EPA 200.7	Spectro ® ICP
Manganese	EPA 200.7	Spectro ® ICP
Iron	EPA 200.7	Spectro ® ICP
Copper	EPA 200.7	Spectro ® ICP
Orthophosphate Phosphorus	EPA 365.2	Beckman® DU 640 Spectrophotometer
Total Phosphorus	EPA 365.4, modified	Perstorp® or Lachat® QuickChem Autoanalyzer
Total Suspended Solids	EPA 160.2	Sartorius® AC210P or Mettler® AT261 analytical balance, oven
Total Solids	SM 2540C	Sartorius® AC210P or Mettler® AT261 analytical balance, oven
Total Volatile Solids	SM 2450G	Sartorius® AC210P or Mettler® AT261 analytical balance, oven, muffle furnace
Total Volatile Solids	EPA 160.4	Sartorius® AC210P or Mettler® AT261 analytical balance, oven, muffle furnace
Potential Hydrogen	EPA 150.1 and EPA 9045A	Accument® AB15 Plus pH meter
Conductivity	EPA 120.1 and EPA 9050A	YSI® 3200 conductivity meter
Aluminum	EPA 200.7	Spectro ® ICP

EPA = Methods for Chemical Analysis of Water and Wastes, March 1983 and version 2, June 1999. There is no difference between EPA methods 200.7 and 6010B. Method 200.7 is a newer version and will yield the same results.

Statistical analysis

Analysis of variance (ANOVA) was performed to examine the treatment effects on nutrients, solids, metals, and other water quality parameters for different sampling profiles (LP, LS, tank supernatant and tank profile). Furthermore, the ANOVA was conducted to investigate the treatment effects among the sampling events and over all sampling events (grand mean). All statistical analyses were performed using SAS software (SAS, 1999) and the Generalized Linear

Model (GLM) procedure. The treatment means were then separated with the Duncan's Multiple Range Test ((Steel and Torrie 1997) at a significance level P of 0.05), if the main treatment effect was significant in the ANOVA.

Results and Discussion

Environmental conditions

Monthly precipitation data for the dairy was provided by the producer and is presented in Fig. 9a. The study area generally received less than four inches of rain per month during the sampling period of August 2007 to March 2008. Higher ambient temperatures were observed during the months of June through September (Fig. 9b) while much lower ambient temperature in December, January, and February were recorded at the study area.

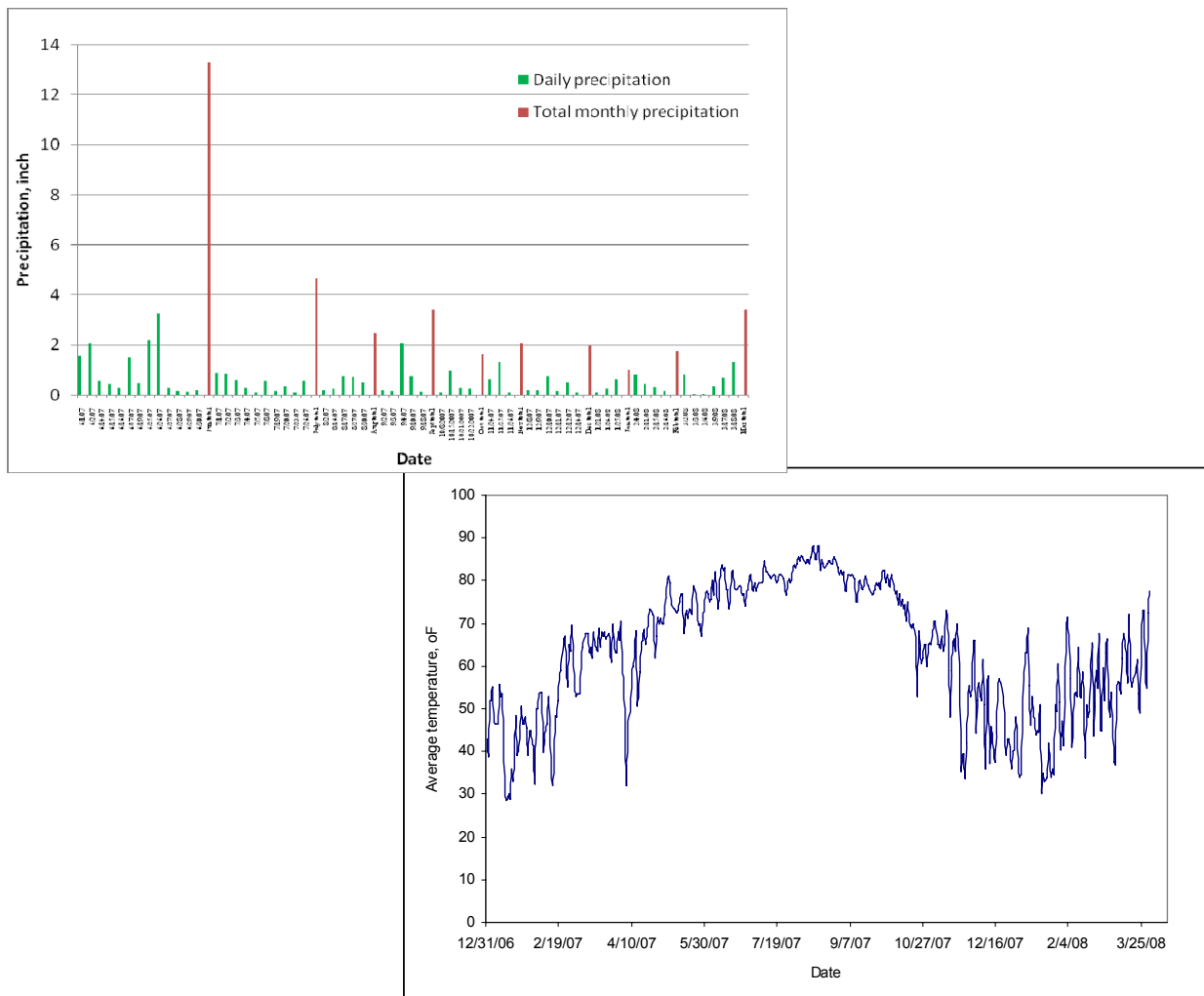
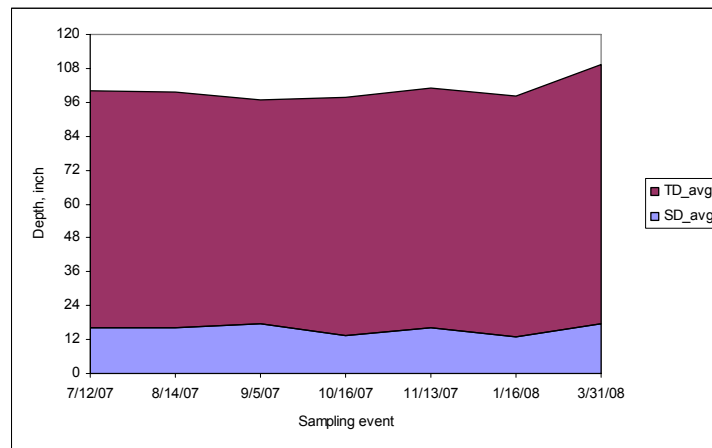


Figure 9b. Recorded ambient temperature trend in the study area.

Lagoon Performance

Sludge Depth

Average TD and SD of the lagoon during each sampling event are shown in Fig. 10. The TD fluctuation was likely due to variations in precipitation, volume of effluent used for irrigation, and evaporation during the monitoring period. The variation in DADS was likely due to variation of settling and re-suspension of solids from microbial activities. Following the first treatment in September 2007, the sludge depth decreased by as much as 20% in October 2007 (Fig. 10). Thereafter, lagoon depths fluctuated slightly, but SD remained lower than the pre-treatment sludge depth (Fig. 10).



**Figure 10. Total and sludge depths of the lagoon
(Note: September, 2007 sampling is the pretreatment depth).**

The likely causes of SD reduction was the loosening of dense sludge from the bottom of the lagoon that came up to the surface due to internal mixing (Zhang et al. 1997) caused by the microbial activities in the lagoon, which was shown during the sampling events. With the tank, it was difficult to measure sludge depth accumulation due to very loose sludge at the bottom of the tank. As a result, no sludge accumulations in the tanks were reported.

Since sludge accumulation is composed of fixed and slowly degradable volatile solids (Chastain et al., 2001), variations in SD are likely due to variation of these solids in this lagoon. In addition, high variability in sludge depth was likely due to internal mixing caused by the microbial activities in the lagoon (Zhang et al. 1997), wind-driven turbulence, gas lift (Reed et

al., 1995), annual cycle of storage, heating, and organic matter accumulation (Hamilton et al., 2006; Westerman et al. 2006). Overall, WTS[®] treatment was somewhat effective in reducing sludge depth by 10% compared to its pre-treatment level. Average SD for this lagoon was 19% of the TD, which is less than 25% of total lagoon depth when rapid sludge accumulation begins (Westerman et al. 2006). Overall TD, DADS, and SD for this lagoon during the monitoring period were 7.06 ft (± 1.01), 5.89 ft (± 0.78), and 1.31 ft (± 0.79), respectively.

Physicochemical characteristics of lagoon

Physicochemical parameters (solids, nutrients, and metals) were analyzed for LP, LS, tank supernatant, and tank profile samples (untreated and treated with bacteria). These parameters were compared between sampling profiles and among sampling events, and averaged over all sampling events (grand mean). All results are the average of composite sample analysis for each sampling event.

pH

The WTS[®] treatment system generally increased pH in the LS as compared to LP (Fig. 11). Significant differences in pH averaged over sampling events were also observed between LS and LP. However, there were no significant differences in pH for LS or LP among sampling events. To begin with, the LS showed slightly higher pH as compared to LP and this difference increased as the treatment process continued. This was likely due to the addition of the WTS[®] solution to the lagoon on a daily basis. Average pH trends in LP and LS are presented in Fig. 11 and average pH for LP and LS were 7.23(± 0.07) and 7.32(± 0.11), respectively.

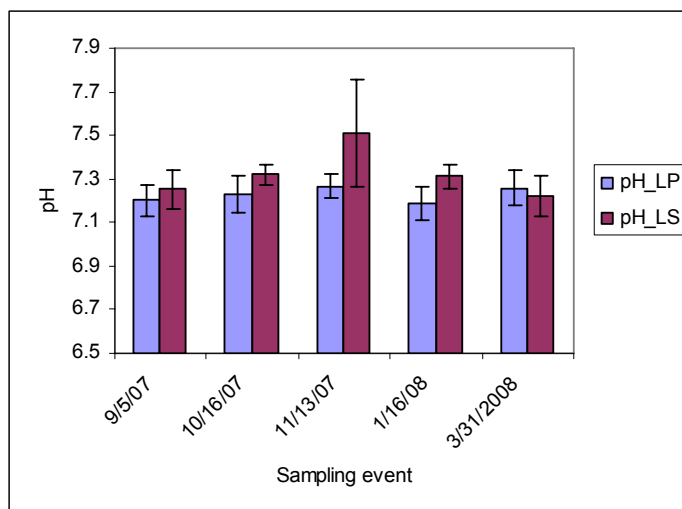


Figure 11. Average pH trends over time for the WTS® treatment. LP: liquid profile, and LS: Liquid supernatant. (Note: September 2007 sampling is the pretreatment sampling.)

Initially, both T1 and T2 tank samples showed similar pH (Fig. 12). Over time, T1 had slightly higher but statistically similar pH to T2 as observed for lagoon pH for LS and LP depths. Slightly increased pH in treated tank T1 samples was likely due to microbial stimulant added to tank T1 and microbial conversion of solids into dissolved solids. Overall, pH of tank profile T1P and T2P were $8.39(\pm 0.87)$ and $7.92(\pm 0.44)$, respectively, and pH of tank supernatant T1S and T2S were $8.68(\pm 0.93)$ and $8.31(\pm 0.57)$, respectively.

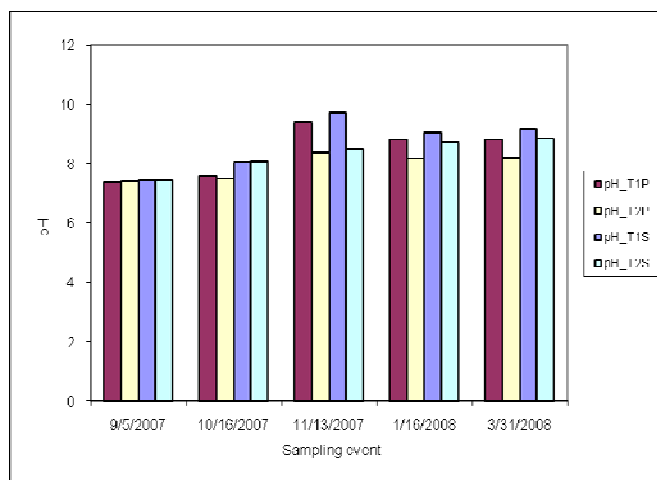
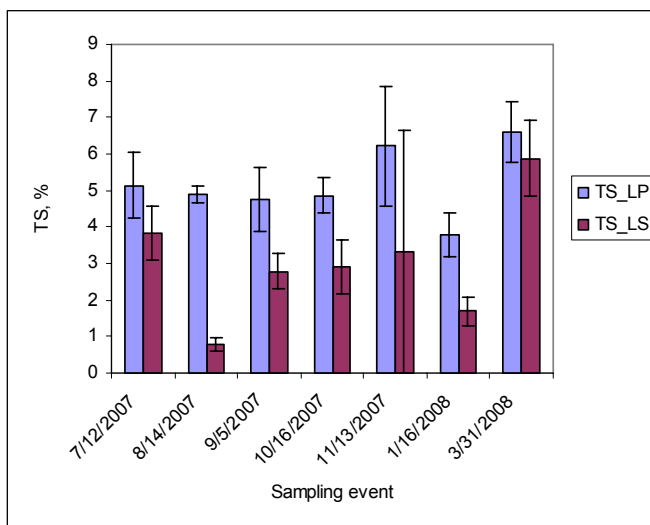


Figure 12. WTS® treatment effects on pH in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Average pH of lagoon (7.28 ± 0.10) was lower than that of the tanks (8.32 ± 0.30), since new flush water was added to the lagoon on a daily basis diluting lagoon wastewater. On the other hand, tanks were filled with flush water at one time and evaporation losses of water from tanks were not compensated with additional water contributing to relatively higher pH in tanks compare to the lagoon. Since pH of the medium profoundly affects the growth of microorganism, slightly higher pH in tanks might slow down the microbial activities and may increase volatilization loss of nutrients. All pH values as received from TIAER lab are listed in tables I through III in Appendix-A.

Solids

Average TS concentrations during each sampling event are shown in Fig. 13 and the overall concentration of TS in LP and LS are listed in Table 3. All solids concentrations as received from the TIAER lab are listed in tables I through III in Appendix-A. Following treatment, TS in both LS and LP increased slightly throughout the monitoring period except during the January 2008 sampling. This may be due to microbial treatment loosening the sludge from the bottom and allowing it to mix with the liquid surface (Zhang et al., 1997) as a result of microbial activities in the lagoon. Overall, TS in LP increased by 9% when averaged over sampling events, whereas TS values in LS increased notably (40%). However, no significant differences in TS were observed when TS was compared between pre-treatment and post-treatment samples within LP and LS profiles. Significant differences in TS were observed between LP and LS, which was expected.



**Figure 13. WTS[®] treatment effects on total solids (TS). LP: liquid profile, LS: Liquid supernatant.
(Note: July – Sept. 2007 sampling are the pre-treatment sampling.)**

Table 3. pH, TS, TSS, TDS, TVS and TFS for lagoon samples averaged over sampling events

Parameter	Sampling location			
	LP		LS	
	Pre-trt	Post-trt	Pre-trt	Post-trt
pH	7.20b*±0.07	7.23b±0.07	7.25a±0.1	7.34a±0.16
Total solids (TS), %	4.29a±1.06	4.99a±1.70	2.21b±1.16	3.32b±2.15
Total suspended solids (TSS), %	4.04a±1.13	3.84a±1.67	1.97b±1.17	2.55b±1.69
Total dissolved solids (TDS), %	0.14b±0.16	0.93a±0.70	0.81a±0.69	0.73a±0.79
Total volatile solids (TVS)	2.92±0.55	3.40±0.92	1.70±0.31	2.10±1.47
Total fixed solids (TFS)	1.83a±0.31	1.93a±0.55	1.08b±0.17	1.25b±0.72

*Pre-trt and post-trt means within a row and profile followed by different letters are significantly different at $P \leq 0.05$ according to Duncan multiple range tests.

Average TS content in tank profiles and tank supernatants during each sampling event are shown in Fig. 14. TS in both treated and untreated tank profiles and tank supernatants decreased slightly and followed a trend similar to each other. No significant differences in TS were observed between treated and untreated tank in any profiles. Decrease in TS from both profiles was likely due to intrinsic microbial activities, but not due to WTS[®] treatment since the differences between treated and untreated tank remained similar. Significant differences in TS were observed between tank supernatant and tank profile within each tank as expected (Fig. 14). Overall, no significant reductions in TS were observed in lagoon or tank environments as a result of WTS[®] treatment.

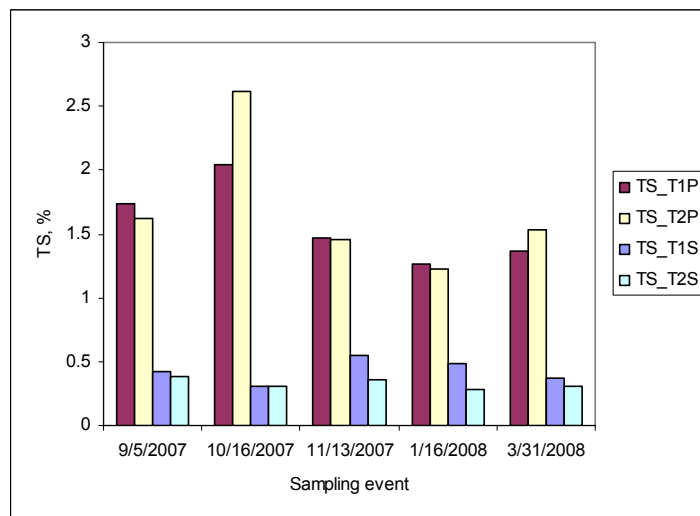


Figure 14. WTS® treatment effects on total solids (TS) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Average TS in both tanks was initially lower than that of the lagoon because the tanks were filled with lagoon wastewater pumped at a shallow depth. Compared with lagoon response, TS concentration in the tank profiles decreased over time (Fig. 14), while TS concentration increased slightly in lagoon profile (Fig. 13). This difference was due to differences in waste loading, microbial activities and light intensity between two conditions. For example, light intensity will be greater at shallow water depth in tanks than the lagoon, and as a result under tank conditions, photosynthetic bacteria will dominate and influence microbial activities (Sund et al. 2001). Overall, no significant reduction in TS was observed in lagoon or tank environments.

The majority of TS concentration increase in the lagoon profile samples occurred when temperatures were favorable for enhanced microbial activity that loosens sludge from the lagoon bottom. As a result, an increase in TS was observed due to internal mixing caused by increased microbial activities in the lagoon.

Overall, average TS for LP and LS (Table 3) were slightly greater than TS concentrations observed by Mukhtar et al. (2004), Barker et al. (2001; cited in Mukhtar et al., 2004), and Converse and Karthikeyan (2004). Solids concentration in LS was also slightly higher (2.4 to 2.6%) than the typical 1% found in the supernatant of most anaerobic dairy lagoons. This higher TS content in LS for this lagoon might be a result of higher solids loading than other lagoons as

well as loosening of sludge due to treatment. This could contribute to greater sludge accumulation if the lagoon is not managed properly.

TSS in LP followed a trend similar to TS, but in LS the TSS concentration decreased gradually following WTS[®] treatment except for in March (Fig. 15). Pre-treatment TSS concentration in LP showed little variation as compared to the TSS concentration post treatment (Fig. 15). Overall, TSS concentration in LP was reduced by 5% when averaged over pre-treatment concentration. In LS, pre-treatment TSS values fluctuated; however, following treatment TSS concentration, reduced gradually until January 2008 sampling, but then increased notably (131%) during the March 2008 sampling. Overall, TSS concentration in LS was increased by 29% when averaged over pre-treatment concentration. For this lagoon, TSS concentration was 86% and 79% of the TS for LP and LS, respectively. Therefore, variability of TS concentration in LP and LS for this lagoon was apparently due to variation in TSS concentrations. Suspended solids can settle on the bottom of lagoon or float on the surface of the lagoon and can affect the lagoon's performance. Figure 15 indicates that this treatment system was not effective in reducing TSS from LP, but reduced TSS somewhat from LS during the monitoring period.

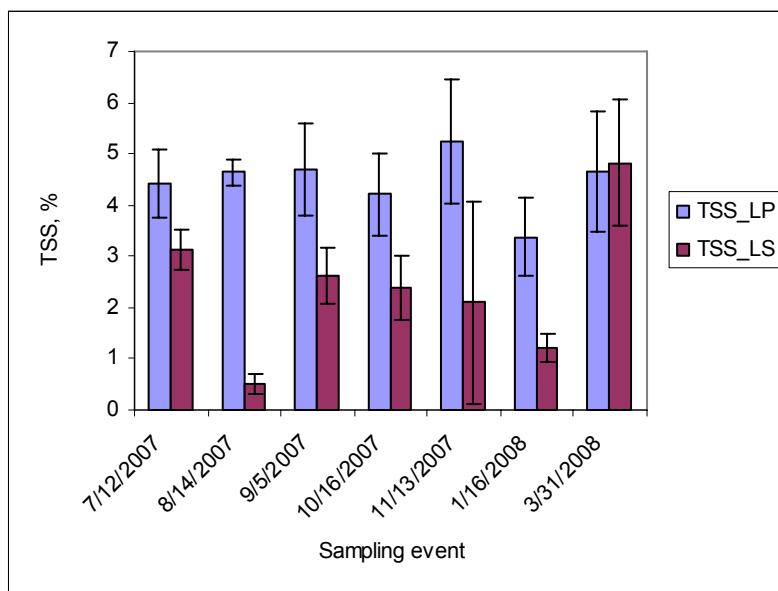


Figure 15. WTS[®] treatment effects on: Total suspended solids (TSS). LP: liquid profile, LS: Liquid supernatant. (Note: July - September 2007 samplings are the pre-treatment sampling.)

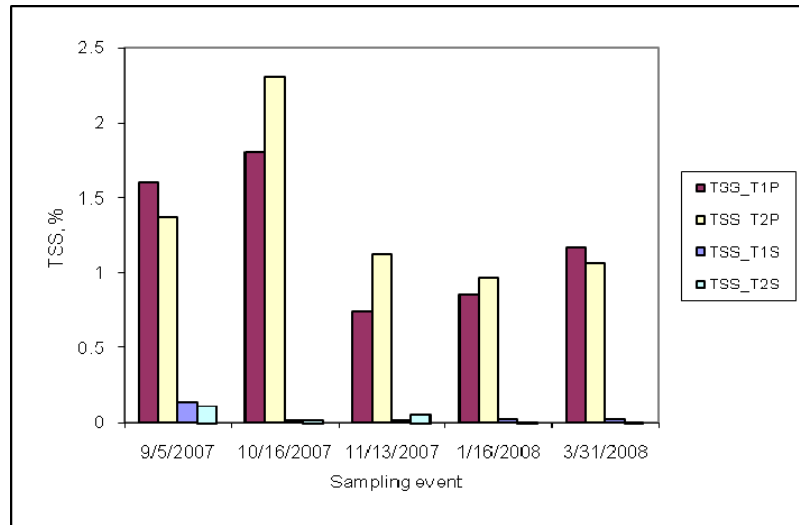
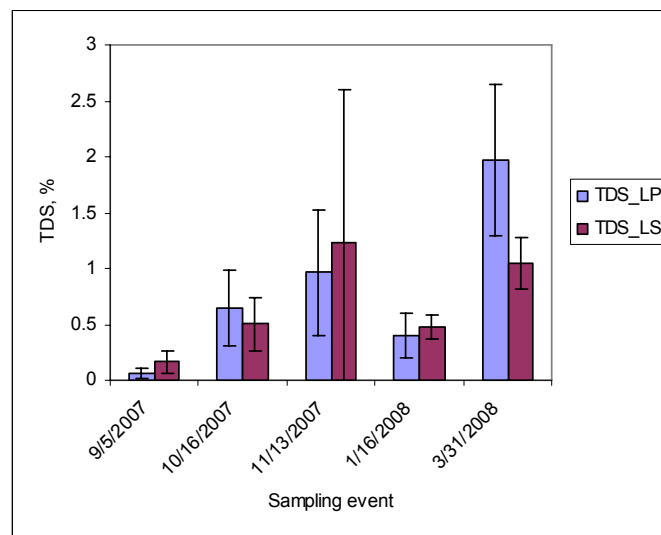


Figure 16. WTS® treatment effects on total suspended solids (TSS) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Over time TSS in both treated and untreated tank samples decreased and followed a trend similar to each other (Fig. 16). Similar TSS reduction in both treated and untreated tank samples was likely due to naturally occurring microbial uptake of organic matters. Although the treated tank showed slightly higher TSS to begin with as compared to the untreated tank, as the treatment continued, the treated tank profile T1P had lower TSS compared to the untreated tank profile T2P (Fig. 16). A similar trend was observed in the tank supernatant samples. This TSS difference in tank profile and tank supernatant between treated and untreated tanks was likely due to WTS® microbial treatment, although TSS difference between treated and untreated tanks was not statistically significant. In future efforts to assess the effectiveness of this treatment system, it might be necessary to monitor the pre- and post-treatment lagoon and tank samples for an extended period of time.

The TS and TSS concentrations of LP were significantly greater than those of LS (Table 3). Average TSS in the LP was higher than LS since suspended solids degrade slowly and remain suspended in the entire LP. In addition, accumulated dead and degraded bacterial mass at the bottom of lagoon might contribute to increased solids content for LP. A similar trend was observed for the tanks.

TDS are easily degradable organic matter and a measure of total materials that are dissolved in water. There were no significant differences in TDS concentration between LP and LS or among sampling events. However, following microbial treatment of the lagoon, TDS concentration both in LS and LP showed an increasing trend except in January (Fig. 17). The TDS concentration in the LP was 19% higher than that of LS, which was likely due to conversion of suspended solids into dissolved solids by the microbes (Zhu et al. 2000) throughout the lagoon profile. Overall, TDS levels in LP and LS increased post-treatment due to microbial activity suggesting that the treatment may not be effective in reducing soluble nutrients from wastewater.



**Figure 17. WTS[®] treatment effects on: a) Total dissolved solids (TDS). LP: liquid profile, LS: Liquid supernatant; IR: Irrigation effluent.
(Note: July - September 2007 samplings are the pre-treatment sampling.)**

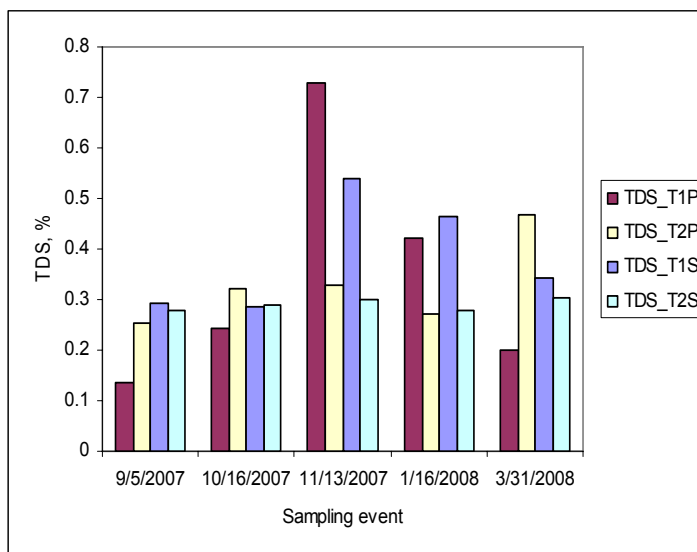


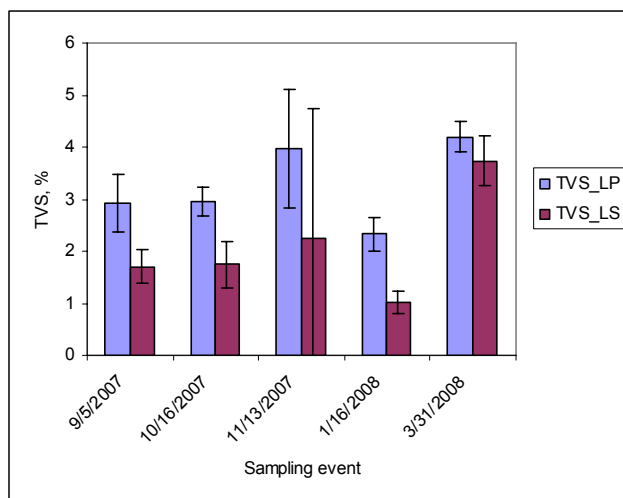
Figure 18. WTS® treatment effects on total dissolved solids (TDS) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

A trend similar to lagoon TDS was also observed in the tank profile samples except in March (Fig. 18). In the tank supernatant, concentration of TDS in the treated tank samples increased towards the end of the monitoring period. TDS concentration was similar throughout the monitoring period of the untreated tank. No significant differences in TDS were observed between treated and untreated tanks, and any observed difference in TDS between the treated and untreated tank was likely due to WTS® treatment effect.

Overall, TDS/TS ratio was relatively higher in LS (0.22) than that of LP (0.15), implying that microbes are more active in the liquid supernatant at converting suspended solids into dissolved solids as compared to the entire profile. Conversely, TDS/TS ratio in the tank profile for the treated and untreated tanks was 0.22 and 0.19, respectively, while they were 0.9 and 0.88 in the tank supernatant for the treated and untreated tanks, respectively.

TVS data are presented in Fig. 19. TVS levels followed a trend similar to TS (Fig. 13) and their concentration increased as the treatment process continued and did not show significant reduction until January 2008. Overall, TVS concentration in LP increased slightly (15%) and TVS in LP constituted 63% of TS. Total volatile solids in LS responded similarly to LP and no

significant reduction (39%) was noticed until January 2008, but TVS concentration increased by 118% in March 2008. Overall, TVS in LS was increased by 28%, which accounted for 69% of TS. Variation in TVS was likely due to variation in the rate and extent of microbial biodegradation of organic compounds and variation of TVS composition (Wilkie 2005) in flushed water added to the lagoon.



**Figure 19. Total volatile solids (TVS) trend over time for the WTS[®] treatment. LP: liquid profile, LS: Liquid supernatant.
(Note: July - September 2007 sampling are the pre-treatment sampling.)**

Following the first treatment, TVS in the TP for both treated and untreated tanks increased, thereafter they reduced gradually (Fig. 20). Overall, TVS reduction in tank profile of the treated and untreated tanks was 26% and 1% while TVS concentration in the tank supernatant was reduced by 54% and 48% in treated and untreated tanks, respectively. In both cases, differences between treated and untreated tanks were not significant.

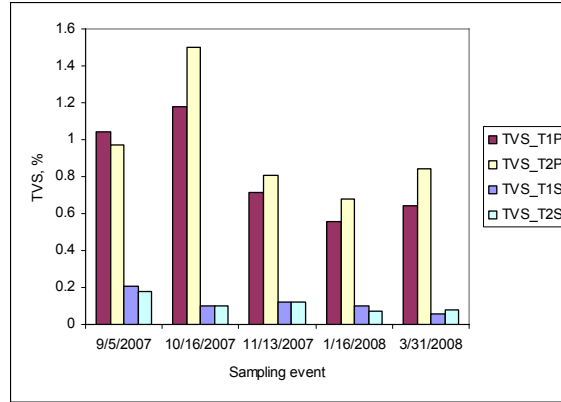


Figure 20. WTS® treatment effects on total volatile solids (TVS) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Total fixed solids (TFS) in LP increased gradually until November 2007 and thereafter started to decrease. Ultimately no significant reduction of TFS was noticed in LS until November 2007 (Fig. 21). The overall increase of TFS in LP was insignificant (<2%), but it reduced in LS by 11%. Over the sampling period, TFS in the tank profiles increased slightly in the treated and untreated lagoon. However, the treated tank yielded a slightly higher TFS concentration (Fig. 22). Significant differences were observed in TFS concentrations between tank profile and tank supernatant samples within treated and untreated tank samples.

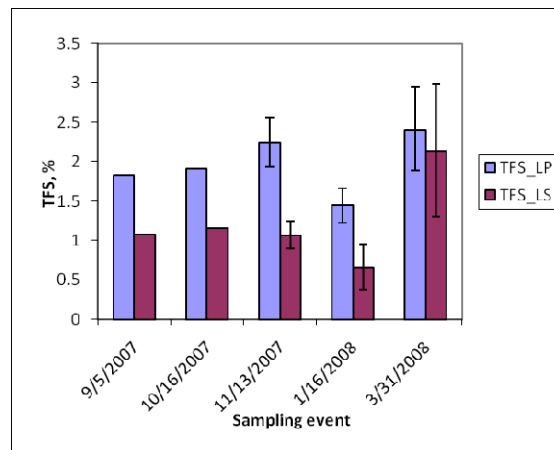


Figure 21. Total fixed solids (TFS) trend over time for the WTS® treatment. LP: liquid profile, LS: Liquid supernatant. (Note: July - September 2007 sampling are the pre-treatment sampling.)

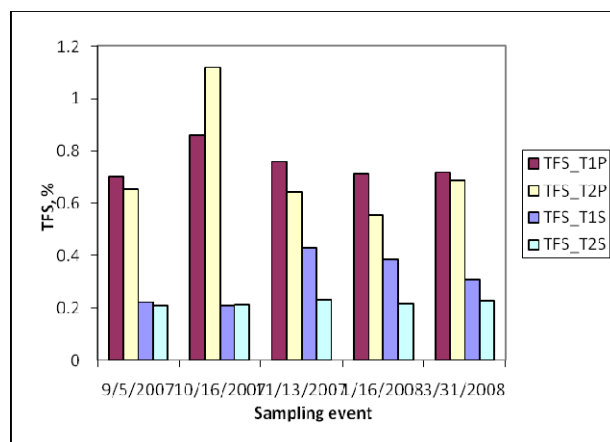
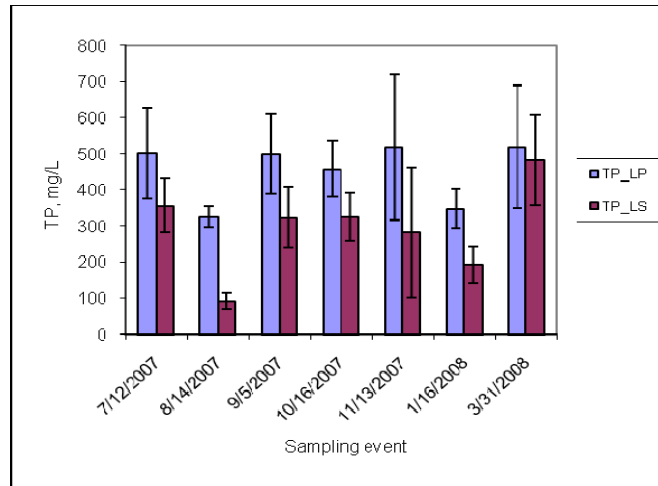


Figure 22. WTS® treatment effects on total fixed solids (TFS) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Typically, TFS is neither chemically reactive nor biologically degradable and theoretically it should stay unchanged (Zhu et al. 2000). For this lagoon, TFS fluctuation suggests that variability in sludge depth was partly due to variation of the solids. Both TDS and TFS for LP were greater than those from LS, although there were no statistically significant differences between LS and LP. Differences were likely due to internal mixing (Zhang et al. 1997) caused by the microbial activities in the lagoon.

Nutrients

Average total P (TP) in LP and LS for each sampling event are presented in Fig. 23 and the concentration averaged over all sampling events is presented in Table 4. The TP in LP was always higher than that in LS for both pre- and post-treatment events (Fig. 23). However, the concentration in both LP and LS fluctuated considerably during the entire sampling period. Overall, no significant reduction in TP was observed but average TP increased about 25% and 4% in LP and LS profiles, respectively, as compare to pre-treatment concentration.



**Figure 23. Total phosphorus (TP) trend over time for the WTS[®] treatment on Total P. LP: liquid profile, LS: Liquid supernatant.
(Note: July - September 2007 sampling are the pre-treatment sampling.)**

As expected, higher TP concentration in LP (Table 4) was likely due to higher TS and TSS concentrations for the LP as compared to LS (Table 3). In addition, degraded microbial cells accumulate at the bottom of the lagoon and runoff water may contribute to increased TP concentration in LP. In this study, no quantitative or qualitative assessment of runoff water additions to the lagoon was conducted, therefore the effects of runoff on the lagoon can not be quantified.

Following the first treatment in September 2007, TP concentration in both tank profiles increased slightly in October 2007, thereafter TP concentration decreased gradually below the pre-treatment concentration (Fig. 24). Overall, TP concentration in the treated tank profile (T1P) decreased by 18%, but increased by 2% in the untreated tank profile samples (T2P). The increase in TP in the untreated tank profile may be due to drastic increase in TP during October 2007 sampling and the reason is unknown. Conversely, TP concentration reductions in treated (T1S) and untreated tank supernatant (T2S) samples were 60% and 55%, respectively. This suggested that the differences in TP reduction between treated and untreated samples were due to treatment effects, whereas reductions of TP in untreated tank samples were likely due to intrinsic microbial activities.

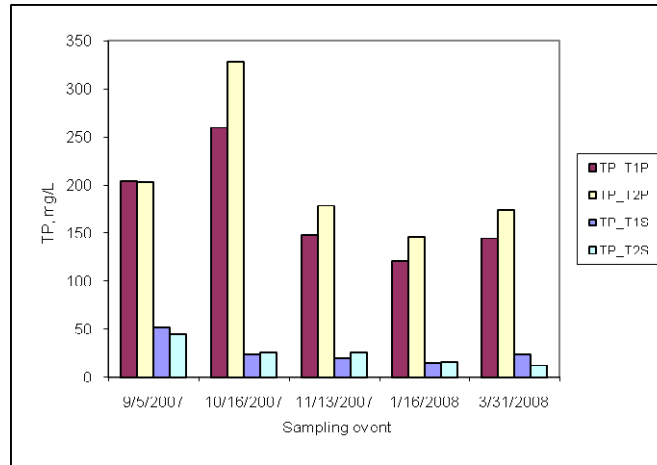


Figure 24. WTS® treatment effects on total phosphorus (TP) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Table 4. Average TP, SRP, and K concentration (mg/L) for lagoon effluent samples averaged over all sampling events

Parameter ¹	Sampling location			
	LP		LS	
	Pre-trt	Post-trt	Pre-trt	Post-trt
Total phosphorus (TP)	385a [*] ±129	397a±185	231b±118	310b±147
Soluble reactive phosphorus (SRP)	11.37a±5.4	13.95a±7.0	10.85a±4.4	13.22a±3.6
Total Kjeldahl nitrogen (TKN)	1666b±642	1258a±405	1323b±258	1029c±399
Nitrate-Nitrite Nitrogen (NNN)	0.06a±0.03	0.34a±0.78	0.06a±0.05	0.08a±0.04
Potassium (K)	404b±29	505a±63	357b±5.9	456a±65

^{*} Pre-trt and post-trt means within a row and profile followed by different letters are significantly different at $P \leq 0.05$ according to Duncan multiple range tests.

¹ parameter is in mg/L

A weak correlation was observed between TP and TS ($R^2=0.37$) and TSS ($R^2=0.27$) in the LP profile. The relatively weak correlation in LP between TS and TP was unexpected, as P typically shows strong association with solids. Conversely, TP was strongly tied with TS ($R^2=0.86$) and TSS ($R^2=0.91$) in the LS (Fig. 25). McFarland et al. (2003) found that TP is partially tied to TSS. In the lagoon, 80% of the TS were TSS in the LS profile and might have contributed to the high correlation with TP. Therefore, without measuring the sludge's P content, the reduction of P from the entire profile due to treatment cannot be unequivocally determined.

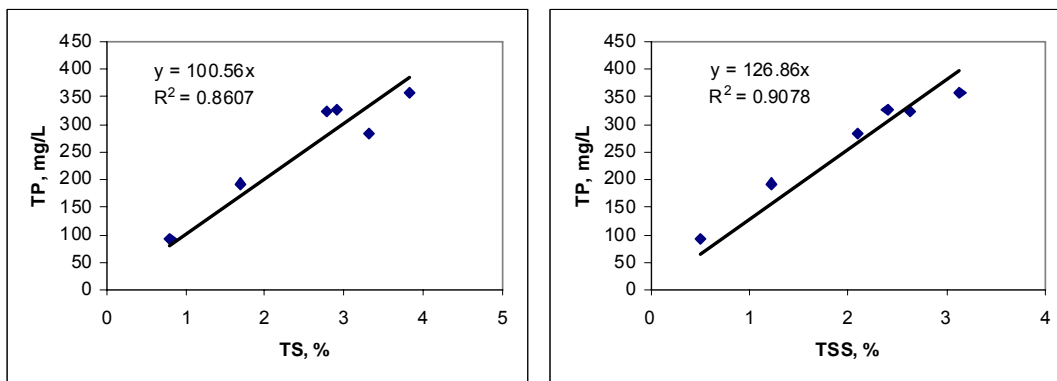


Figure 5. Relationship between TP vs. TS and TP vs. TSS for LS profile.

Average SRP levels in LP and LS during each sampling event are presented in Fig. 26. Pre-treatment SRP varied widely with no definite trend in both cases, but following microbial treatment, SRP concentrations for these sampling locations increased gradually. This increase in SRP concentration was likely due to loosening of sludge from the lagoon bottom as well as runoff water contributions of unknown quality and quantity to the lagoon. The SRP concentrations averaged over all sampling events (combined pre- and post- treatment) in LS and LP were (11.20 ± 3.13) and (11.54 ± 3.44) , respectively, and statistically similar (Table 4).

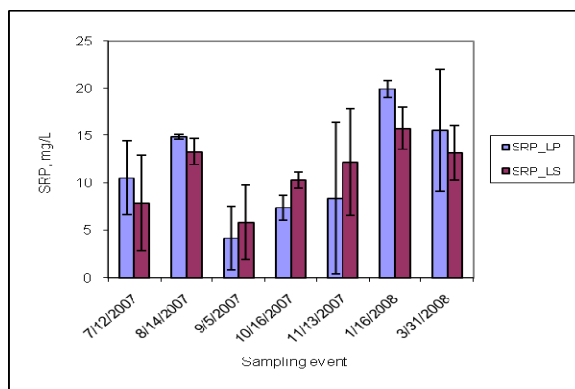


Figure 26. Soluble reactive phosphorus (SRP) concentration trends over time for the WTS® treatment. LP: liquid profile, LS: Liquid supernatant. (Note: July - September 2007 sampling are the pre-treatment sampling.)

A similar SRP increasing trend was observed in tank profile and supernatant samples and the treated tank had higher SRP concentrations than that of untreated tank samples (Fig. 27). This was most likely due to greater TDS in the treated tank samples. Overall, no significant differences in SRP were observed between treated and untreated tank samples. Researchers (Converse and Karthikeyan 2004) have indicated that loosening of the settled solids from the lagoon bottom may cause them to rise to the upper profile, carrying the P associated with them, which might increase SRP. Despite this, variations of solids show little effect on SRP concentration for this lagoon as also observed by other researchers (Vanotti et al. 2007).

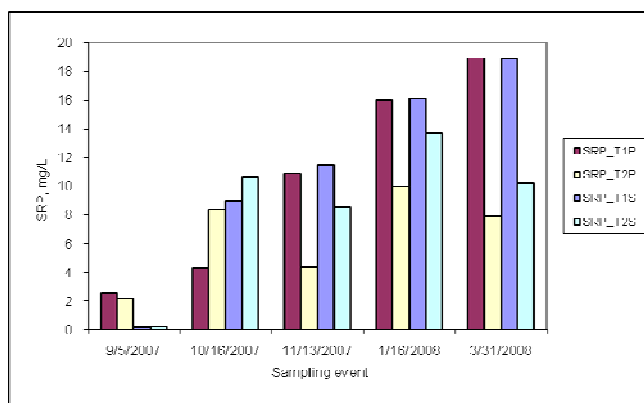


Figure 27. WTS® treatment effects on soluble reactive phosphorus (SRP) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Following the pre-treatment sampling in September 2007, post-treatment TKN in LP fluctuated and decreased slightly. The TKN also decreased slightly in LS throughout the monitoring period (Fig. 28). Significant differences in TKN concentrations were observed between pre- and post-treatment for both LP and LS profiles (Table 4), however no significant differences were observed among sampling events within each profile. Overall, TKN reduction in LP and LS were 29% and 19%, respectively. The reduction of TKN concentration in LP and LS were likely due to a combination of ammonia volatilization (Higgins et al. 2004), flush water added to the lagoon (Scotford et al. 1998), and treatment effects.

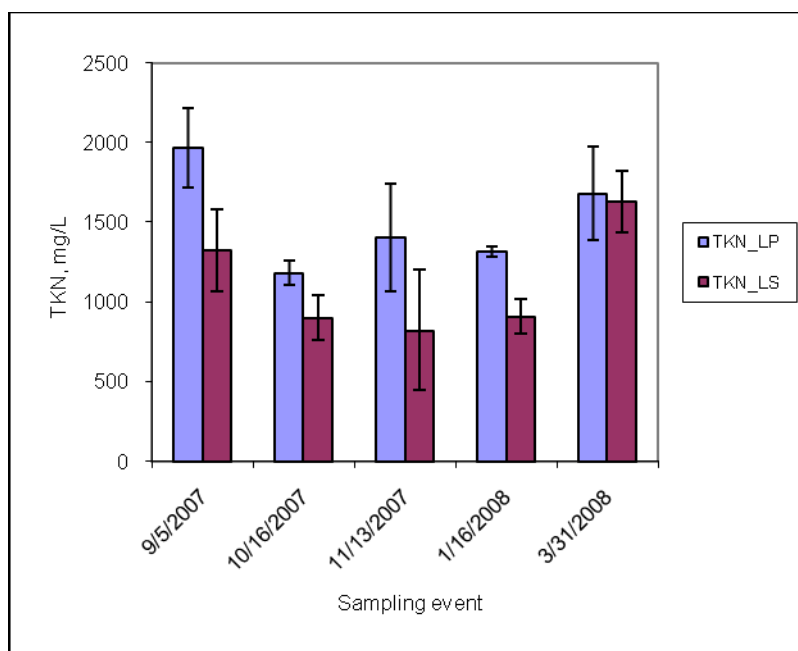


Figure 28. Total Kjeldahl nitrogen (TKN) concentration trends over time for the WTS® treatment. LP: liquid profile, LS: Liquid supernatant.
(Note: September 2007 sampling is the pre-treatment sampling.)

The TKN concentration in both the untreated and treated tank profile and tank supernatant samples reduced considerably following pre-treatment sampling in October 2007. These concentrations reduced further in November 2007 and then remained fairly constant till the end of sampling period (Fig. 29). The TKN reduction rate in the tank profile for the treated tank was slightly greater (58%) than that of untreated (47%). Similarly, TKN reductions in tank supernatant in treated and untreated tanks were 88% and 86%, respectively. However, no significant differences in TKN reduction were observed between treated and untreated tank samples (both liquid profile and supernatant). This implies that reduction of TKN under tank conditions was not due to WTS® treatment, but may be due to ammonia volatilization losses.

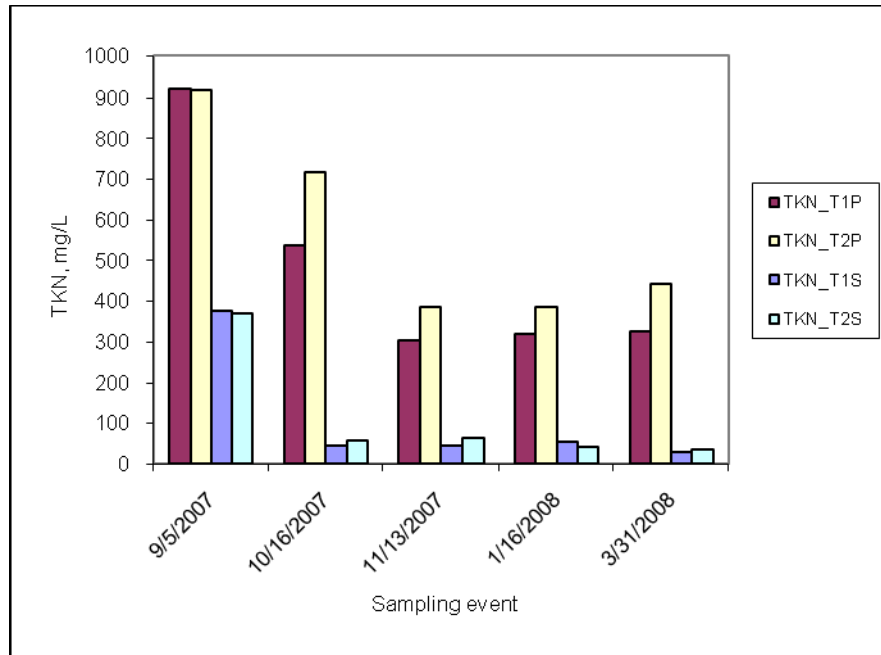


Figure 29. WTS® treatment effects on Total Kjeldahl nitrogen (TKN) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

Average NNN concentrations for LP and LS are presented in Fig. 30. Following pre-treatment sampling in September 2006, NNN concentration increased tremendously in the LP during October 2007 before gradually decreasing to near pre-treatment concentrations. A similar trend was also observed in LS, however, the magnitude was much smaller than the LP. The NNN concentration increases in these profiles were likely due to ammonia diffusing upward from the bottom of the lagoon profile and converted into nitrate via nitrification process (Nealson 1997). Overall, no significant differences in NNN concentration were observed between LP and LS (Table 4) and this treatment was not effective in reducing NNN concentrations since 60%-70% of NNN is soluble (Bicudo et al. 1999) and it is difficult to reduce soluble concentration.

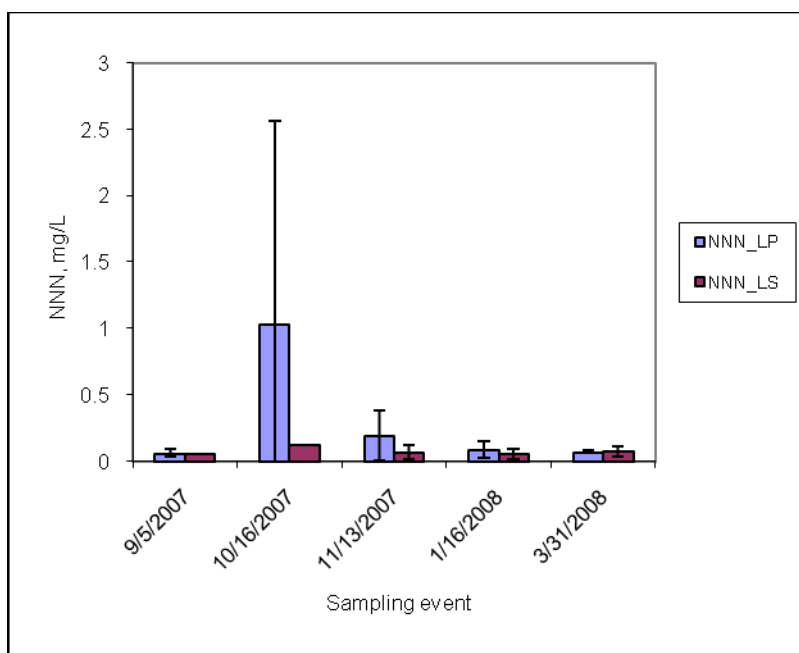


Figure 30. Nitrite-Nitrate Nitrogen (NNN) concentration trends over time for the WTS[®] treatment. LP: liquid profile, LS: Liquid supernatant. (Note: September 2007 sampling is the pre-treatment sampling.)

Similarly, NNN concentrations both in treated and untreated tank profile and tank supernatant samples increased considerably over the time (Fig. 31). The differences between tank profile and tank supernatant as well as pre-treatment and post-treatment samples were not significant. This increase in NNN concentrations in the treated tank samples was likely due to conversion of ammonia nitrogen into nitrite and nitrate nitrogen. In addition, evaporation loss of water might also contribute to greater NNN concentration in both tank samples.

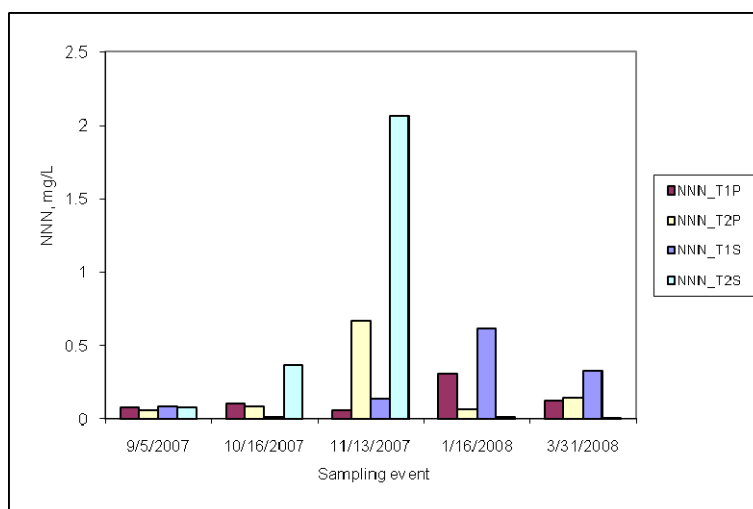


Figure 31. WTS® treatment effects on Nitrate-Nitrite Nitrogen (NNN) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

The K concentration in LP was always higher than LS for both pre- and post-treatment sampling events and showed an increasing trend following microbial treatment throughout the monitoring period (Fig. 32). The K concentration followed a trend similar to TDS (Fig. 17). This increase in K concentrations was likely due to runoff water contribution and variations in flush water added to the lagoon. In addition, dissolved solids might also contribute to increased K concentration since K is highly soluble in water (Gustafson et al. 2007). There were significant differences in K concentrations between pre- and post-treatment LP and LS samples (Table 4). It is apparent that this microbial treatment was not effective in reducing K concentrations of any profiles since K is highly soluble.

K concentrations in both treated and untreated tanks also increased over time (Fig. 33). The increase in K concentration in tank samples was likely due to evaporation loss and contributions from increased dissolved solids as a result of Ks high solubility. Overall, K concentrations in the treated tank were slightly higher compared to the untreated tank and were due to WTS® treatment effects.

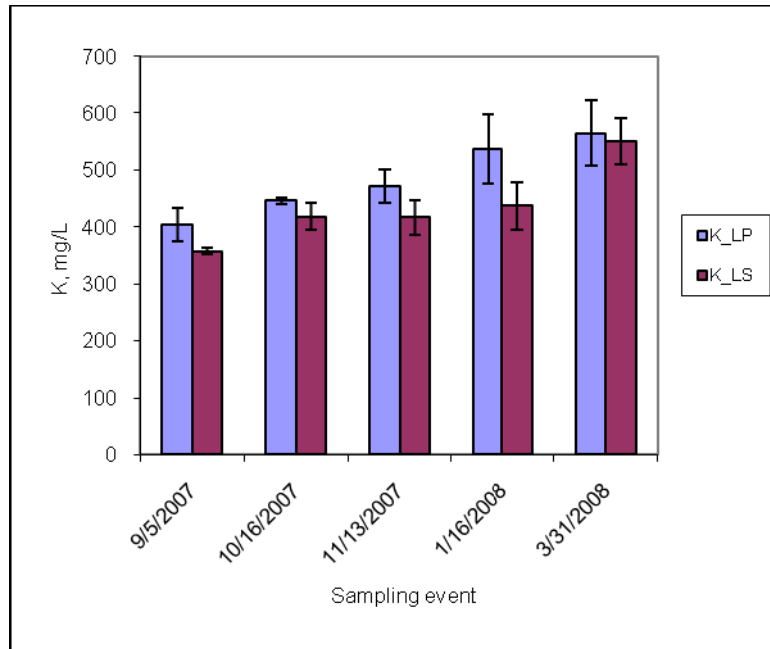


Figure 32. Potassium (K) concentration trends over time for the WTS® treatment.
 LP: liquid profile, LS: Liquid supernatant.
 (Note: September 2007 sampling is the pre-treatment sampling.)

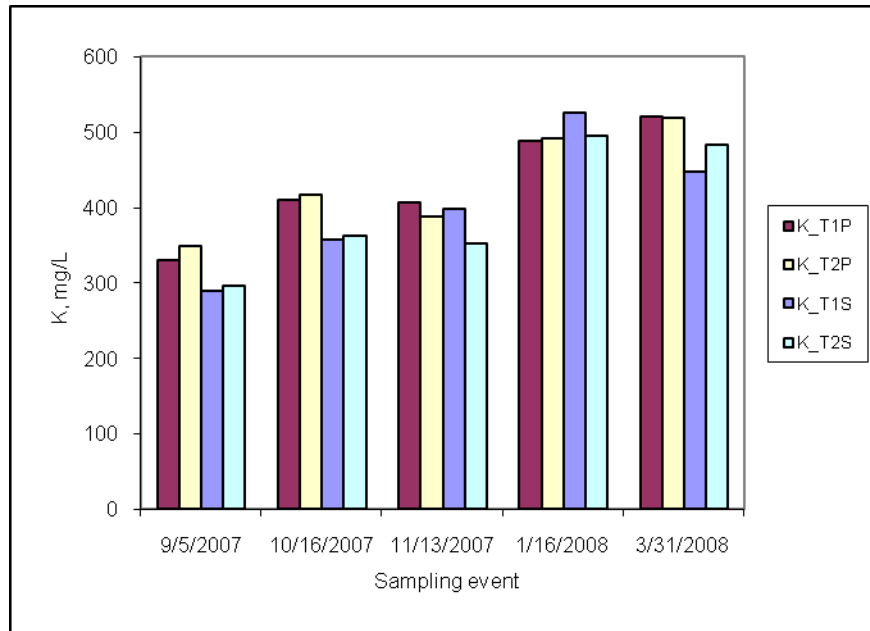


Figure 33. WTS® treatment effects on potassium (K) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2.
 (Note: September 2007 sampling is the pre-treatment sampling.)

Nutrient data analyses suggest that WTS[®] treatment was not effective in reducing TP from any of the lagoon profiles. In the tank environment, notable TP concentration reduction trends were observed for the treated and untreated tank samples with the treated tank showing slightly greater reduction trends. This implies that the treatment was somewhat effective in reducing TP. Conversely, SRP and K in LP and LS increased, while TKN decreased slightly. A similar trend was also observed for these parameters under tank sampling. This implies that the reduction in nutrients under lagoon and tank environment were likely due to combination of WTS[®] treatment and naturally occurring microbial uptake of nutrients, settling of solids, and flush water added to the lagoon. However, without the accurate measurement of sludge nutrient content, especially P in lagoon sludge, it was difficult to ascertain that the reduction or increase of nutrients from these profiles was likely due to settling of solids or WTS[®] treatment effects. All nutrients concentration as received from TIAER lab are also listed in Tables I through III in the Appendix. Typically, three chemical quality parameters indicate the effectiveness of a wastewater treatment system such as biological oxygen demand (BOD), suspended solids, and TP (VanLoon and Duffy 2000). Suspended solids and TP were both monitored in this study and showed insignificant variation between pre-treatment and post-treatment events. Therefore, this treatment system was not very effective in reducing phosphorus and other nutrients from the lagoon effluent, especially soluble parameters.

Metals

Minerals in dietary amount are required for normal growth and reproduction of animals (NRC 2001). The metals content in animal manure is largely a reflection of metals concentration in the feed animals consumed and the efficiency of feed conversion by animals (Nicholson et al. 1999). Following microbial treatment, Al, Ca, Cu, and Fe concentrations in both LP and LS decreased slightly. Mg concentrations in the LP remained same as pre-treatment concentration throughout the monitoring period and its concentration reduced slightly in the LS. In case of Mn, its concentration fluctuated in the LP but gradually decreased in the LS except at the end of sampling period. Either a similar or slight reduction in Al concentrations in both LP and LS samples were observed (Fig. 34). A similar trend was also observed for the concentrations of other metals for different sampling events in LP and LS, except Mg (Tables 5 and 6). Overall, no

notable reduction in concentrations of Al, Ca, Cu, Fe, and Mn were observed from any of these profiles following microbial treatments (Table 7).

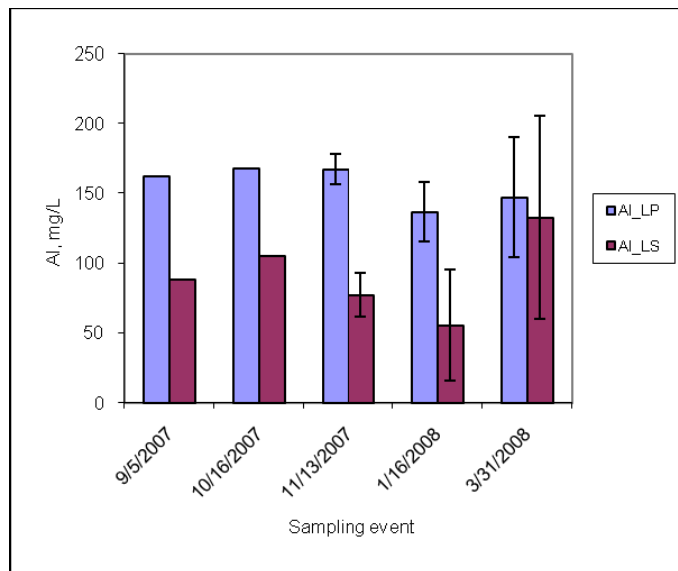


Figure 34. Aluminum (Al) concentration trends over time for the WTS® treatment.
LP: liquid profile, LS: Liquid supernatant.
(Note: July – September 2007 sampling are the pre-treatment sampling.)

Al concentrations in both treated and untreated tank profile and tank supernatant increased in October 2007, thereafter they followed a similar decreasing trend till the end of the sampling events (Fig. 35). A similar trend was observed for Ca and Fe. In addition, samples collected from the treated tank showed higher reduction rates for these metals as compared to the untreated tank and was likely due to treatment effects. Irrespective of treatment, these metals' concentrations decreased from both tank profiles which were likely due to intrinsic microbial metabolic activities. Conversely, Cu, Mg and Mn concentration in the tanks fluctuated over time and was likely due to the environmental conditions in the tank. Microorganisms can promote mineral formation or degradation based on environmental situation (Ehrlich 1997). These variations in metal concentration in both lagoon and tank environmental situation were also likely due to the variations in feed composition, which was not explored because it was beyond the scope of this study.

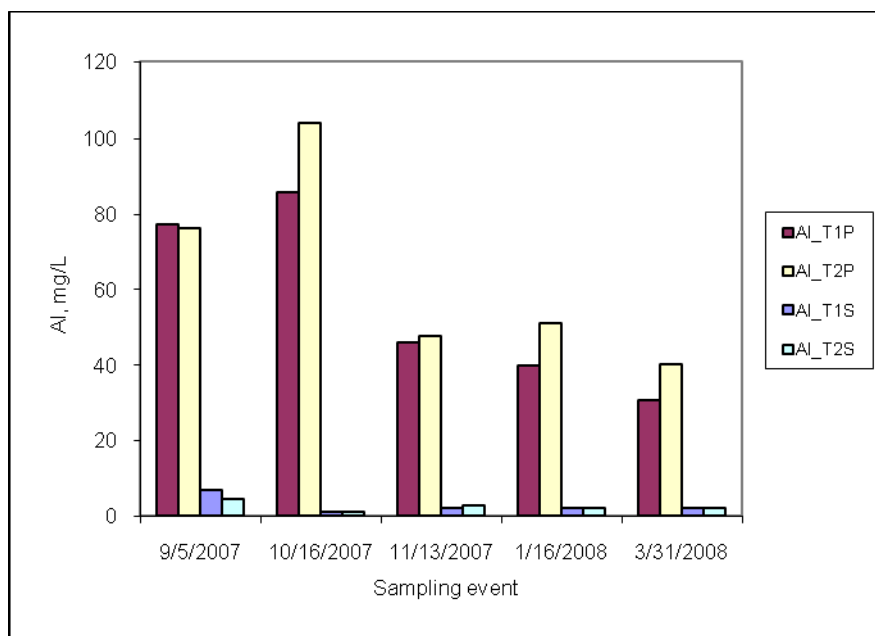


Figure 35: WTS® treatment effects on Aluminum (Al) in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: Sep, 2007 sampling is the pre-treatment sampling.)

Table. 5. Average concentration of Calcium (Ca), Copper (Cu) concentration for LP and LS at different sampling events

Date	Ca (mg/L)		Cu (mg/L)		Fe (mg/L)	
	LP	LS	LP	LS	LP	LS
09/05/07	3153a±647	1980ab±272	50.6ab±15.0	30.2b±7.7	171a±26.8	82.8a±12.4
10/16/07	3263a±408	2120ab±448	48.5ab±10.4	30.1b±10.3	169a±17	85.6a±48.2
11/13/07	2980a±938	1450ab±1123	46.7ab±19.7	23.1b±21.7	162a±50	70.2a±70.1
1/16/08	2730a±672	1250b±488	38.3b±13.5	14.9b±7.7	138.4a±45	44.1a±13.1
3/31/08	3627a±1011	3333a±893	73.7a±11.4	68.4a±9.0	197a±60	174b±50

*Averages within a column followed by different letters are significantly different at $P \leq 0.05$ according to Duncan multiple range tests.

Table. 6. Average concentration of magnesium (Mg), manganese (Mn) and sodium (Na) for LP and LS at different sampling events

Date	Mg(mg/L)		Mn(mg/L)		Na(mg/L)	
	LP	LS	LP	LS	LP	LS
09/05/07	366a±55.4	265b±24	16.5ab±3.1	10.0b±1.6	288ab±11	260b±4
10/16/07	356a±83	304a±40	13.1ab±1.0	8.4b±2.7	277b±95	321a±14
11/13/07	366a±71	251a±86	16.7ab±6.3	8.9b±7.7	313ab±9	293a±8.5
1/16/08	359a±63	232a±42	12.0b±2.6	5.2b±1.9	378a±41	321a±20
03/31/08	421a±93	398a±75	23a±8.4	20.4a±7.2	313ab±21	317a±21

Averages within a column followed by different letters are significantly different at $P \leq 0.05$ according to Duncan multiple range tests

Table 7. Average metals concentration (mg/L) for LP and LS sampling locations averaged over all sampling events

Parameter ¹	Sampling location			
	LP		LS	
	Pre-trt	Post-trt	Pre-trt	Post-trt
Aluminum (Al)	162a±11	156a±33	89a±16	93a±49
Calcium (Ca)	3153a±647	3150a±761	1980a±272	2038a±1085
Copper (Cu)	171a±27	51.80a±18.38	30a±8	34.14a±24.30
Iron (Fe)	171a±27	167a±45	83a±12	93a±66
Manganese (Mn)	16.53a±3.06	16.20a±6.46	9.96a±1.60	10.71a±7.64
Magnesium (Mg)	366a±55	376a±72	265a±24	296a±87
Sodium (Na)	288a±11	320a±59	260b±59	313a±19

* Averages within a row followed by different letters are significantly different at $P \leq 0.05$ according to Duncan multiple range tests

Nicholson et al. (1999) reported that the mean Cu concentration in dairy cattle slurry collected from commercial farms in England and Wales was 4.73 mg/L (62.3 mg/kg dm; dry matter 7.6%). Ullman and Mukhtar (2007) reported Cu concentrations in dairy lagoons in central Texas in the range of 8.1-19.2 mg/L depending on management practices applied at the specific dairy. In this study, average Cu concentration in LP and LS was 46.01 ± 14.64 and 24.56 ± 11.86 mg/L, respectively, which is much higher than reported elsewhere. Cu concentration in manure is related to Cu added as a supplement to feed (Li et al. 2005). In general, manures will contain higher Cu concentration if feeds contain higher Cu levels (Nicholson et al. 1999). In this study, feed composition was not analyzed, however, average concentrations of metals (i.e., Ca, Mg, Fe, etc.) in the lagoon were similar to those values reported by Mukhtar et al. (2004). The values reported for Ca and Fe were two and nine times higher, respectively, than the values reported by

Mukhtar et al. (2004), but Fe concentration was comparable with North Carolina's reported values. All metals concentrations as received from TIAER lab are also listed in Tables I and II of Appendix A.

Conductivity

The average conductivity in LP and LS is presented in Fig. 36. The microbial treatment (WTS[®]) appeared to cause a slight increase in EC levels until the end of the experiment. However, no significant differences in conductivity were observed between LP and LS samples. On the other hand, conductivity and K demonstrated a strong correlation in LP ($R^2 = 0.99$) and LS ($R^2 = 0.71$) samples. A strong correlation was also observed between conductivity and Na in LP ($R^2 = 0.86$), however a weak correlation for the two parameters was observed in LS ($R^2 = 0.44$). These results showed a good agreement with the findings of Scotford et al. (1998), who observed high correlation ($R^2 = 0.80$) between K and EC.

The treated tank (T1) samples resulted in slightly higher conductivity values than the untreated tank samples (Fig. 37), which was likely due to treatment effects.

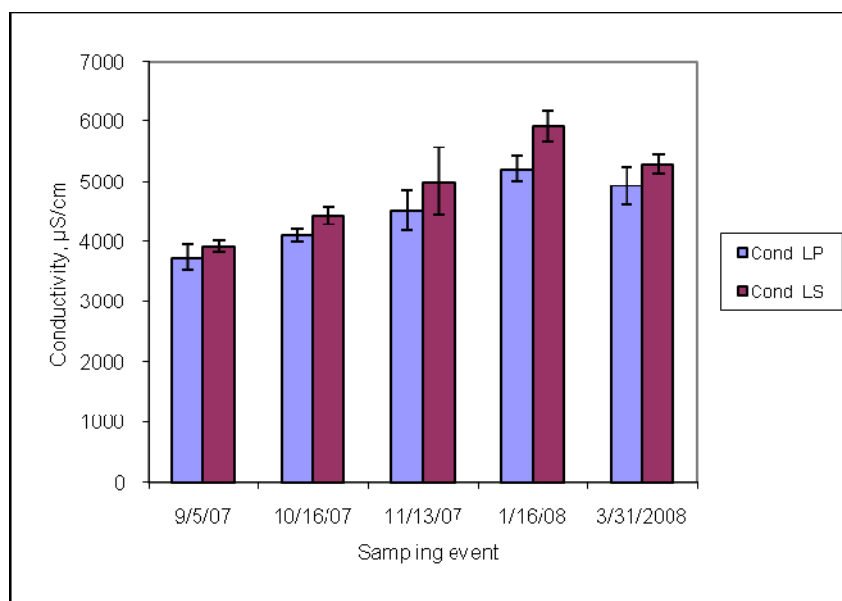


Figure 36. Conductivity trends over time for the WTS[®] treatment. LP: liquid profile, LS: Liquid supernatant.

(Note: July - September 2007 sampling are the pre-treatment sampling.)

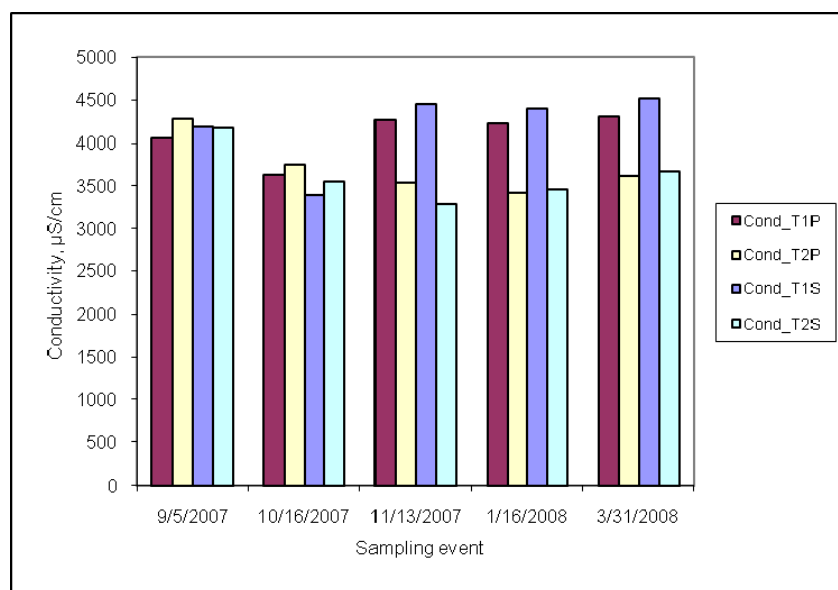


Figure 37. WTS® treatment effects on conductivity in tank profiles. T1P: tank profile in treated tank T1, T2P: tank profile in untreated tank T2, T1S: liquid supernatant in treated tank T1, T2S: liquid supernatant in untreated tank T2. (Note: September 2007 sampling is the pre-treatment sampling.)

While statistically similar, the average conductivity for LS (4929 ± 239 µS/cm), was slightly higher than LP (4503 ± 233 µS/cm). Safley et al. (1993) reported that EC values of 8000 µS/cm can inhibit bacterial population in a livestock operation's treatment lagoon. In this lagoon, EC was lower than this suggested threshold value. All conductivity values as received from TIAER lab are listed in Tables I through III in Appendix A.

Treatment Costs

Costs to implement this lagoon treatment method varied based on the daily amount of manure and wastewater added to the lagoon, the existing lagoon capacity and sludge depth, prior wastewater treatment (e.g., pretreatment of flushed manure for solids separation before it flows to the lagoon), lagoon depth, and the number of lagoon cells in the wastewater management system. Treatment costs will also vary with the type of manure alley cleaning system used, such as flushing or vacuuming. The following cost matrix was provided by the technology provider.

Table 8. Cost to treat a lagoon with WTS[®] microbial treatment

Herd size	Unit cost (\$/cow/month)	\$/cow/year
1000	0.50	6
1001-7000		
>7001		

Based upon the information in Table 8, for this 600-head dairy, the total cost to treat the lagoon was estimated at \$2,100 for a 7-month period or \$0.50/cow/month.

Conclusion

The WTS[®] treatment was somewhat effective in reducing sludge depth by 10% compared to its pre-treatment level. This reduction of sludge depth was due to microbial treatment. This treatment system significantly increased lagoon pH in the LS as compared to LP. Similar to lagoon pH, over-time treated tank T1 indicated slightly higher but statistically similar pH as compared to untreated tank T2 in both tank profiles. There was no significant reduction in TS either in lagoon or tank environments due to WTS[®] treatment. Overall, lagoon TSS concentration in LP was reduced by 7% when the post-treatment levels were compared to the pre-treatment levels, whereas, this reduction was 9% in the LS. Over time, TSS in both treated and untreated tank samples decreased and followed trends similar to lagoon TSS concentrations. Following microbial treatment of the lagoon, TDS concentrations both in LS and LP increased, although these differences were not significant. Overall, TDS concentration in the LS was 13% higher than that observed in LP.

There was no significant reduction in TP between treated or non-treated lagoon sampling profiles. Conversely, TP concentration in treated tank profile was reduced by 17% and increased by 2% in the untreated tank profile. On the other hand, the TP values declined in the treated and untreated tank supernatant samples by 60% and 55%, respectively. These differences in TP reduction between treated and untreated samples were due to WTS[®] treatment effects. There were no significant differences in SRP concentrations between LP and LS samples from the lagoon, and they showed increasing trend overtime. A similar increasing trend for SRP was observed in both treated and untreated tank samples. However, the treated tank showed higher SRP concentrations than that of the untreated tank samples as a result of greater TDS in tank supernatant. The TKN in LP and LS decreased by 29% and 19%, respectively, and a larger TKN reduction was observed in the tank profile (60% and 47% in treated and untreated tank profile samples, respectively) and tank supernatant (88% to 86% in treated and untreated tank supernatant samples, respectively) samples as compared to lagoon samples. The K concentration and conductivity increased in both lagoon and tank sample profiles throughout the monitoring period. Overall, there were no significant reductions in TS, TP, and SRP between treated and non-treated lagoon sampling profiles. The main purpose of this study was to observe the

effectiveness of WTS® in reducing P and other substances from lagoon effluent to be applied to WAFs. Therefore, this treatment system was not effective in reducing phosphorus and other nutrients from the lagoon effluent, especially soluble parameters.

Challenges

Tanks were used to mimic the repeatability of lagoon treatment with microbes and to get additional information on treatment effectiveness. Due to evaporation losses, it was difficult to maintain a consistent TS and TP sampling depth in the tanks. It was possible to continue sampling, although TP sampling depth varied due to water losses from tanks. It remains a challenge to obtain replicated data on treatment effectiveness in outdoor environmental conditions under a tank environment. It is apparent that microbial treatment was more effective in the lagoon supernatant than the entire profile but, without accurate assessment of pre- and post-treatment sludge characteristics, it is premature to conclude how effective the treatment was in reducing nutrient, metal, and solids levels in the lagoon. The foremost challenge is to collect and monitor the lagoon sludge sample for an extended period of time prior to, during, and after treatment to determine solids, nutrients and metal content of the lagoon that will enable a determination to be made regarding the effectiveness of the applied treatment.

Acknowledgements

This project was supported by the Texas State Soil and Water Conservation Board and EPA-CWA section 319 (h). Ozona[®] Environmental LLC is the technology provider.

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APPENDIX - A

Table I. pH, conductivity, solids, nutrients and metals concentration at different sampling locations and sampling events in LS profiles

Site ID	Date	pH	Cond	NNN	SRP	TP	TKN	TS	TSS	TVS	TDS	TFS	Al	Ca	Cu	Fe	K	Mg	Mn	Na
L1S	9/5/07	7.15	4028	0.031	9.07	240	1090	24500	23200	14800	1310	9700	70.3 ^{SR}	1750 ^{SR}	23.2 ^{SR}	71.9 ^{SR}	353 ^{SR}	241 ^{SR}	8.85	256 ^{SR,D}
L1S	10/16/07	7.28	4607	0.086	10.6	254	760	22900	20600	13800	2320	9100	67.2 ^{SR}	1730 ^{SR}	19.2 ^{SR}	62.8 ^{SR}	408 ^{SR}	272 ^{SR}	7.67	319 ^{SR,D}
L1S	11/13/07	7.37	5490	0.109	16.1	97.4	408	7290	3200	3800	4090	3490	15	409	3.69	15	394	176	1.79	293
L1S	1/16/08	7.25	6150	0.053	13.6	149	809	14200	10400	8990	3800	5210	40.9	841	8.83	33.3	395	194	3.68	302
L1S	3/31/08	7.11	5460	0.085	11.1	395	1600	47400	34400	32100	13000	15300	94.6	2330	58	117	504	314	12.2	293
L2S	9/5/27	7.31	3893	0.031	7.03	323	1280	25700	23000	15700	2710	10000	96 ^{SR}	1910 ^{SR}	28.9 ^{SR}	80.2 ^{SR}	364 ^{SR}	264 ^{SR}	9.23	262 ^{SR,D}
L2S	10/16/07	7.31	4395	0.137	11	384	1040	37100	31200	22400	5880	14700	146 ^{SR}	2610 ^{SR}	39.6 ^{SR}	141 ^{SR}	446 ^{SR}	349 ^{SR}	11.4	336 ^{SR,D}
L2S	11/13/07	7.36	5130	0.063	14.7	297	908	21700	17200	13000	4500	8700	59.8	1300	19	46.5	406	233	7.76	284
L2S	1/16/08	7.34	5660	0.039	15.7	248	1020	21300	15400	12800	5900	8500	72.1	1790	23.6	58.7	479	277	7.35	341
L2S	3/31/08	7.28	5250	0.066	16.5	429	1450	61400	53200	38300	8230	23100	150	3630	73.7	198	569	421	23.5	331
L3S	9/5/27	7.3	3866	0.12	1.42	409	1600	33500	32600	20700	864	12800	99.6 ^{SR}	2280 ^{SR}	38.4 ^{SR}	96.2 ^{SR}	355 ^{SR}	289 ^{SR}	11.8	263 ^{SR,D}
L3S	10/16/07	7.37	4328	0.152	9.34	343	904	27200	20200	16300	6960	10900	102 ^{SR}	2020 ^{SR}	31.6 ^{SR}	53.0 ^{SR}	402 ^{SR}	292 ^{SR}	6.11	308 ^{SR,D}
L3S	11/13/07	7.79	4400	0.037	5.78	454	1150	70600	42400	50600	28200	20000	157	2640	46.6	149	450	345	17	301
L3S	1/16/08	7.35	5980	0.071	18	180	899	15200	10800	8950	4400	6250	53.8	1120	12.2	40.3	438	224	4.57	320
L3S	3/31/08	7.27	5150	0.074	11.9	626	1840	67500	57200	41600	10300	25900	154	4040	73.6	207	578	458	25.5	328

Table II. pH, conductivity, solids, nutrients and metals concentration at different sampling locations and sampling events in LP profiles

Site ID	Date	pH	Cond	NNN	SRP	TP	TKN	TS	TSS	TVS	TDS	TFS	Al	Ca	Cu	Fe	K	Mg	Mn	Na
L1P	9/5/07	7.12	3716	0.044	6.31	399	1770	40100	39800	24600	274	15500	153 ^{SR}	2590 ^{SR}	36.1 ^{SR}	147 ^{SR}	372 ^{SR}	315 ^{SR}	14.2	277 ^{SR,D}
L1P	10/16/07	7.13	4220	2.8	8.71	377	1110	43000	33600	26400	9450	16600	145 ^{SR}	2810 ^{SR}	36.7 ^{SR}	151 ^{SR}	453 ^{SR}	261 ^{SR}	12	322 ^{SR,D}
L1P	11/13/07	7.22	4890	0.151	17.6	335	1100	48200	40400	31000	7800	17200	118 ^{SR}	2000 ^{SR}	26.4	106 ^{SR}	440 ^{SR}	293 ^{SR}	9.47	304 ^{SR}
L1P	1/16/08	7.1	5410	0.157	19.2	302	1350	33900	28000	21700	5900	12200	99.1	2010 ^{SR}	24.8	88.1	470	289	9.34	332 ^{SR}
L1P	3/31/08	7.21	5190	0.084	23	338	1370	56800	37200	38600	19600	18200	104	2540	60.6	130	503	320	13.4	290
L2P	9/5/07	7.25	3963	0.045	5.91	482	1890	45400	44200	27600	1150	17800	159 ^{SR}	3010 ^{SR}	49.6 ^{SR}	167 ^{SR}	410 ^{SR}	358 ^{SR}	15.4	288 ^{SR,D}
L2P	10/16/07	7.27	4078	0.157	7.31	469	1180	50400	43000	30300	7380	20100	187 ^{SR}	3600 ^{SR}	52.4 ^{SR}	170 ^{SR}	445 ^{SR}	402 ^{SR}	13.6	168 ^{SR,D}
L2P	11/13/07	7.25	4440	0.399	4.4	483	1350	57800	52600	35400	5200	22400	188	3070	48	178	476	372	19	315
L2P	1/16/08	7.24	5000	0.071	19.6	409	1320	44500	42600	27000	1900	17500	156	3340	51.7	175	554	409	14.5	393
L2P	3/31/08	7.21	4590	0.066	11.8	546	1730	68900	42400	43100	26500	25800	158	3800	79.4	215	577	440	26.3	320
L3P	9/5/07	7.24	3535	0.093	0.23	620	2250	57200	56800	35400	446	21800	174 ^{SR}	3860 ^{SR}	66.1 ^{SR}	200 ^{SR}	429 ^{SR}	425 ^{SR}	20	299 ^{SR,D}
L3P	10/16/07	7.28	4010	0.132	6.14	530	1260	52500	49600	31800	2850	20700	171 ^{SR}	3380 ^{SR}	56.4 ^{SR}	185 ^{SR}	441 ^{SR}	406 ^{SR}	13.8	340 ^{SR,D}
L3P	11/13/07	7.33	4250	0.032	3.16	736	1770	80300	64400	52500	15900	27800	196	3870	65.7	203	498	434	21.5	321
L3P	1/16/08	7.22	5230	0.03	20.9	336	1280	34900	30800	21200	4100	13700	156	2840	38.3	152	588	380	12.2	410
L3P	3/31/08	7.35	5020	0.06	11.9	675	1950	72500	59600	44100	12900	28400	180	4540	81.2	246	615	502	29.3	330
LP10	9/5/07				14.2	148	753	15800	12000		3850									
L10S	10/16/07				17.6	183	600	18000	15400	11400										
LP10	11/13/07				11.9	73	308	10500	7100		3400									
LP10	1/16/08				24.4	98.7	688	50300	42000	39000	8300	11300								

Table III. pH, conductivity, solids, nutrients and metals concentrations at different sampling locations and sampling events in tank conditions

Site ID	Date	pH	Cond	NNN	SRP	TP	TKN	TS	TSS	TVS	TDS	TFS	Al	Ca	Cu	Fe	K	Mg	Mn	Na
T1S	9/5/07	7.43	4176	0.087	0.14	51.5	375	4270	1340	2070	2930	2200	6.78 ^{SR}	252 ^{SR}	<1.00 ^{SR}	4.74 ^{SR}	289 ^{SR}	132 ^{SR}	<1.00	229 ^{SR,D}
T1S	10/16/07	8.04	3388	< 0.011	9.02	23.9	44.8	3040	170	966	2870	2074	<1.00 ^{SR}	42.3 ^{SR}	<1.00 ^{SR}	<1.00 ^{SR}	357 ^{SR}	83.2 ^{SR}	<1.00	284 ^{SR,D}
T1S	11/13/07	9.73	4450	0.142	11.5	19.9	43.1	5500	120	1220	5380	4280	< 2	52.6	< 2	< 2	397	131	<1	668
T1S	1/16/08	9.03	4390	0.618	16.1	15.3	52.7	4850	200	1000	4650	3850	< 2	123	< 2	< 2	526	158	< 2	920
T1S	3/31/08	9.17	4520	0.333	18.9	23.2	28	3660	216	602	3440	3058	<2.00	52.5	<2.00	<2.00	447	22.2	<2.00	735
T2S	9/5/07	7.44	4173	0.08	0.224	44.2	368	3880	1080	1800	2800	2080	4.53 ^{SR}	230 ^{SR}	<1.00 ^{SR}	3.29 ^{SR}	295 ^{SR}	132 ^{SR}	<1.00	230 ^{SR,D}
T2S	10/16/07	8.07	3555	0.369	10.7	25.5	55.6	3110	200	1010	2910	2100	<1.00 ^{SR}	57.5 ^{SR}	<1.00 ^{SR}	<1.00 ^{SR}	362 ^{SR}	149 ^{SR}	<1.00	284 ^{SR,D}
T2S	11/13/07	8.48	3290	2.06	8.51	25.6	61.8	3570	570	1250	3000	2320	2.82	71.2	< 2	< 2	351	131	<1	239
T2S	1/16/08	8.71	3460	< 0.015	13.7	15.7	39.5	2850	52	712	2800	2140	< 2	49	< 2	< 2	495	175	< 2	389
T2S	3/31/08	8.85	3670	< 0.015	10.2	12.7	33.2	3080	60	800	3020	2280	<2.00	34.2	<2.00	<2.00	482	148	<2.00	299
T1P	9/5/07	7.38	4049	0.078	2.57	204	922	17400	16000	10400	1370	7000	77.3 ^{SR}	1380 ^{SR}	17.2 ^{SR}	57.7 ^{SR}	330 ^{SR}	215 ^{SR}	6.39	248 ^{SR,D}
T1P	10/16/07	7.57	3623	0.105	4.32	259	537	20400	18000	11800	2440	8600	85.8 ^{SR}	1620 ^{SR}	15.2 ^{SR}	47.2 ^{SR}	410 ^{SR}	266 ^{SR}	5.35	299 ^{SR,D}
T1P	11/13/07	9.39	4260	0.064	10.9	148	302	14700	7400	7130	7300	7570	46	702	11.7	40.9	406	180	4.3	665
T1P	1/16/08	8.8	4220	0.31	16	121	319	12700	8500	5580	4200	7120	39.8	749	9.47	29.5	488	203	3.1	852
T1P	3/31/08	8.81	4300	0.128	18.9	144	324	13600	11600	6430	2010	7170	30.5	821	11.7	36.6	520	201	4.47	772
T2P	9/5/07	7.41	4271	0.063	2.17	203	919	16200	13700	9680	2540	6520	76 ^{SR}	1360 ^{SR}	16.4 ^{SR}	53.8 ^{SR}	348 ^{SR}	219 ^{SR}	6.19	255 ^{SR,D}
T2P	10/16/07	7.49	3742	0.09	8.37	328	716	26200	23000	15000	3230	11200	104 ^{SR}	2080 ^{SR}	27.5 ^{SR}	65.8 ^{SR}	417 ^{SR}	289 ^{SR}	7.82	311 ^{SR,D}
T2P	11/13/07	8.38	3540	0.669	4.37	178	386	14500	11200	8080	3300	6420	47.6	848	14	43.7	388	195	5.05	313
T2P	1/16/08	8.16	3420	0.07	9.98	146	385	12300	9600	6760	2700	5540	51	863	11.8	37.4	491	227	3.96	356
T2P	3/31/08	8.2	3620	0.153	7.93	174	442	15300	10600	8450	4680	6850	40.2	1050	15.2	45.2	518	233	5.2	312